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Proceedings, 29th Annual Meeting, Aquatic Plant Control Research Program

**14-17 November 1994
Vicksburg, Mississippi**

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Proceedings, 29th Annual Meeting, Aquatic Plant Control Research Program

**14-17 November 1994
Vicksburg, Mississippi**

Final report

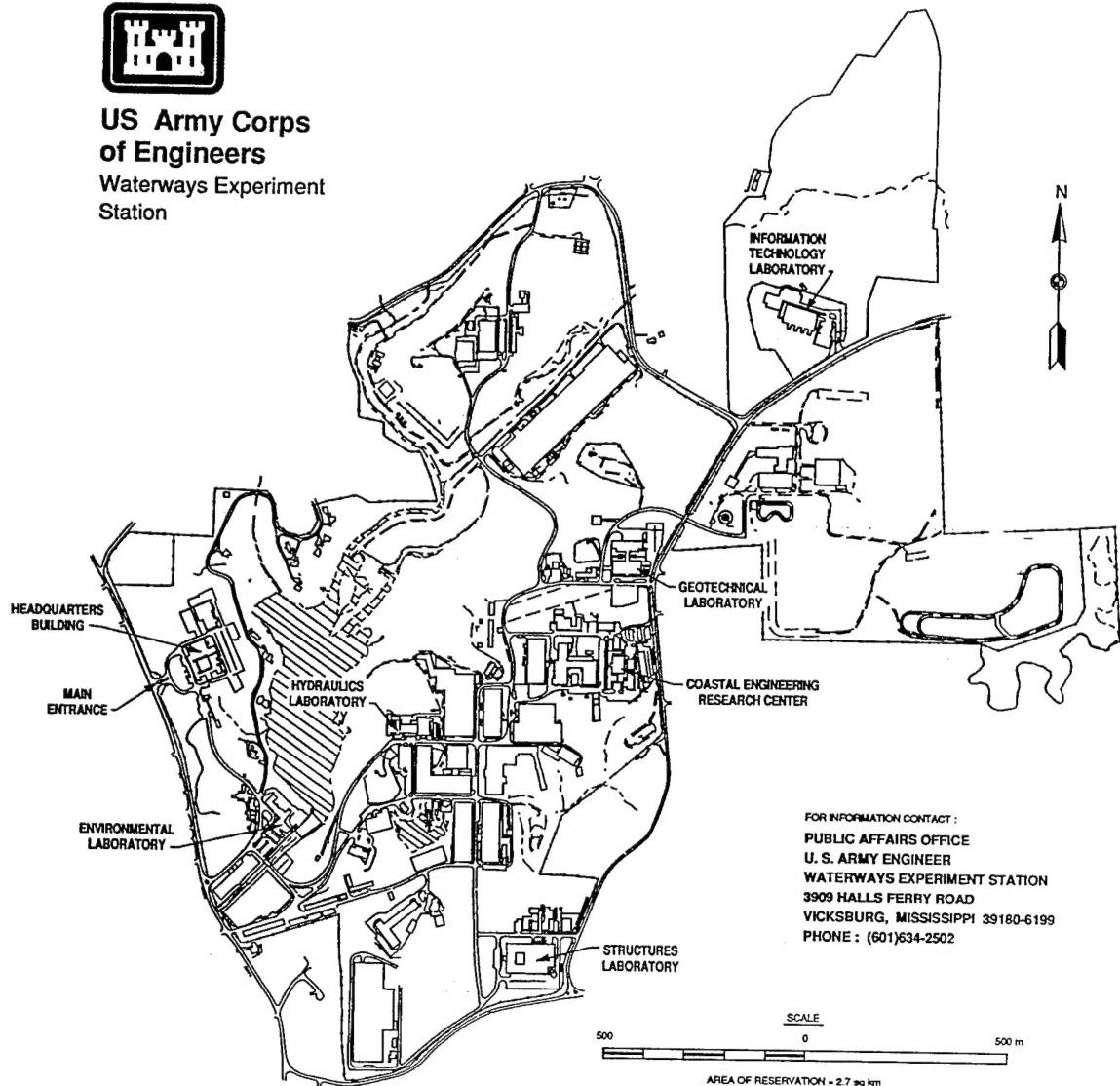
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Station



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Preface

The 29th Annual Meeting of the U.S. Army Corps of Engineers Aquatic Plant Control Research Program (APCRP) was held in Vicksburg, MS, on 14-17 November 1994. The meeting is required by Engineer Regulation 1130-2-412, paragraph 4c, and was organized by personnel of the APCRP, which is managed under the Environmental Resources Research and Assistance Programs (ERRAP) of the Environmental Laboratory (EL), U.S. Army Engineer Waterways Experiment Station (WES), Vicksburg, MS.

The organizational activities were carried out and presentations by WES personnel were prepared under the general supervision of

Mr. J. L. Decell, Manager, ERRAP, EL. Mr. Robert C. Gunkel, Assistant Manager, ERRAP, was responsible for planning the meeting. Dr. John W. Keeley was Director, EL, WES. Ms. Denise White was Program Monitor for the Headquarters, U.S. Army Corps of Engineers.

Ms. Billie F. Skinner, ERRAP, was responsible for coordinating the necessary activities leading to publication.

At the time of publication of this report, Director of WES was Dr. Robert W. Whalin. Commander was COL Bruce K. Howard, EN.

Agenda

Monday, 14 November 1994

- 1:00 p.m. ***Registration***
- 5:00 p.m. Auditorium Lobby
- 1:00 p.m. ***Center for Aquatic Plant Research and Technology***
- 3:00 p.m. ***Advisory Board Meeting***
 ERD Conference Room
- 3:00 p.m. ***Federal Aquatic Plant Management Working Group***
- 5:00 p.m. ERD Conference Room

Tuesday, 15 November 1994

- 7:30 a.m. ***Registration***
- 5:00 p.m. Auditorium Lobby
- 8:00 a.m. ***Poster and Demonstration Session***
- 5:00 p.m. Classroom No. 2 and Auditorium Lobby
- 8:00 a.m. ***General Session***
- 2:00 p.m. Auditorium
- 8:00 a.m. Call to Order and Announcements
* Robert "Bob" C. Gunkel, Waterways Experiment Station (WES),
 Vicksburg, Mississippi
- 8:05 a.m. Welcome to WES
* Robert W. Whalin, Director, WES, Vicksburg, Mississippi
- 8:15 a.m. Center for Aquatic Plant Research and Technology
* J. Lewis Decell, WES, Vicksburg, Mississippi
- 8:45 a.m. Annual Report - Aquatic Plant Control Operations Support Center (APCOSC)
* Wayne T. Jipsen, USAE District, Jacksonville, Florida
- 9:00 a.m. Use of Economics in Evaluation of Aquatic Plant Control
* Jim E. Henderson, WES, Vicksburg, Mississippi

- 9:15 a.m. Growth and Mortality Estimates for Grass Carp in Lakes Moultrie and Marion (32738)
James P. "Phil" Kirk, WES, Vicksburg, Mississippi
- 9:30 a.m. Integrated Control Using Herbicides and Pathogens (32953)
Michael "Mike" D. Netherland, WES, Vicksburg, Mississippi
- 9:45 a.m. Break

Chemical Control Technology

Presiding: Kurt D. Getsinger, WES, Vicksburg, Mississippi

- 10:15 a.m. Chemical Control Technology Development: Overview
Kurt D. Getsinger, WES, Vicksburg, Mississippi
- 10:30 a.m. Herbicide Application Technique Development for Flowing Water:
Summary of Accomplishments (32354)
* Kurt D. Getsinger, WES, Vicksburg, Mississippi
- 10:45 a.m. PGR Update: Flurprimidol (32578)
* Carole A. Lembi, Purdue University, West Lafayette, Indiana
- 11:00 a.m. Herbicide Concentration / Exposure Times: Effects on Nontarget Plants (32352)
* Susan L. Sprecher, WES, Vicksburg, Mississippi
- 11:15 a.m. Herbicide Delivery Systems: Evaluation of New Formulations (32437)
* Michael "Mike" D. Netherland, WES, Vicksburg, Mississippi
- 11:30 a.m. Development of Endothall CR Formulations (32437)
* David Sisneros, Bureau of Reclamation, Denver, Colorado
- 11:45 a.m. Species-Selective Control of Eurasian Watermilfoil Using Triclopyr (32841)
* R. Michael "Mike" Smart, WES, Lewisville, Texas
- 12:00 noon LUNCH
- 1:00 p.m. Field Evaluation of Triclopyr in Lake Minnetonka: Overview (32404)
* Kurt D. Getsinger, WES, Vicksburg, Mississippi
- 1:15 p.m. Triclopyr/Dye Dissipation in Water: Lake Minnetonka (32404)
* Alison M. Fox, Center for Aquatic Plants, University of Florida, Gainesville, Florida
- 1:30 p.m. Changes in a Submersed Plant Community Following Treatment with Triclopyr:
Lake Minnetonka (32404)
* John D. Madsen, WES, Lewisville, Texas
- 1:45 p.m. Phenology of Eurasian Watermilfoil: Determination of Control Points (32441)
* John D. Madsen, WES, Lewisville, Texas
- 2:00 p.m. ADJOURN
- 2:00 p.m. **USAE Division / District Working Session**
- 5:00 p.m. Main Conference Room
- 2:00 p.m. **Joint Agency Guntersville Project Meeting**
- 5:00 p.m. EPED Conference Room

3:00 p.m. **Center for Aquatic Plant Research and Technology**
- 5:00 p.m. **Aquatic Herbicide Working Group**
ERD Conference Room

Wednesday, 16 November 1994

8:00 a.m. **Poster and Demonstration Session**
- 2:30 p.m. Classroom No. 2 and Auditorium Lobby

8:00 a.m. **General Session**
- 2:30 p.m. Auditorium

Biological Control Technology

Presiding: Alfred "Al" F. Cofrancesco, WES, Vicksburg, Mississippi

- 8:00 a.m. Overview of Biocontrol Research
* Alfred "Al" F. Cofrancesco, WES, Vicksburg, Mississippi
- 8:15 a.m. Application of Foundational Research
* Ted D. Center, U.S. Department of Agriculture
(USDA), Agricultural Research Service (ARS), Ft. Lauderdale, Florida
- 8:30 a.m. Hydrilla Defense Genes: Implications in Biocontrol
* Stewart L. Kees, WES, Vicksburg, Mississippi
- 8:45 a.m. Foreign Surveys and Quarantine Studies for Biocontrol of Eurasian Watermilfoil
and Hydrilla (32730)
* Gary R. Buckingham, USDA, ARS, Gainesville, Florida
- 9:00 a.m. The Use of Pathogens for the Management of Hydrilla and Eurasian Watermilfoil
(32202, 32200, 32735)
* Judy F. Shearer, WES, Vicksburg, Mississippi
- 9:15 a.m. European Surveys for Pathogens of Eurasian Watermilfoil (32863)
* Harry Evans, International Institute of Biological
Control, Silkwood Park, Berkshire, United Kingdom
- 9:30 a.m. Current Status of the Hydrilla Biocontrol Program (31799, 32730, 32734)
* Michael "Mike" J. Grodowitz, WES, Vicksburg, Mississippi
- 9:45 a.m. BREAK
- 10:15 a.m. Assessment of Impact of Biocontrol Agents of Hydrilla (31799, 32730)
* Gregory "Greg" Wheeler, University of Florida, Ft. Lauderdale, Florida
- 10:30 a.m. The Effects of Temperature on the Leaf-Mining Flies, *Hydrellia pakistanae*
and *H. balciunasi* (32734)
* Ramona H. Warren, WES, Vicksburg, Mississippi
- 10:45 a.m. Surveys for Biocontrol Agents of *Trapa natans*
* Robert "Bob" Pemberton, USDA, ARS, Ft. Lauderdale, Florida
- 11:00 a.m. Transferring Information on the Use of Biocontrol Technology (32864)
* Michael "Mike" J. Grodowitz, WES, Vicksburg, Mississippi

Ecology of Aquatic Plant Technology

Presiding: John W. Barko, WES, Vicksburg, Mississippi

- 11:15 a.m. Overview of Ecological Studies: Environmental Effects
* John W. Barko, WES, Vicksburg, Mississippi
- 11:30 a.m. Effects of Submersed Macrophytes on Water Quality (32405)
* William "Bill" F. James, WES, Spring Valley, Wisconsin
- 11:45 a.m. Water Movement in Submersed Macrophyte Beds (32405)
* Craig S. Smith, WES, Spring Valley, Wisconsin
- 12:00 noon LUNCH
- 1:00 p.m. Factors Contributing to the Decline of Eurasian Watermilfoil
in TVA Reservoirs (32805)
* Craig S. Smith, WES, Spring Valley, Wisconsin
- 1:15 p.m. Potential Sources of Mineral Nutrition for Submersed Macrophytes
in Riverine Systems: Results of Initial Investigations (32805)
* Sara J. Rogers, National Biological Survey, Onalaska, Wisconsin
- 1:30 p.m. Transport, Deposition, and Fate of Submersed Macrophyte Propagules
in the Potomac River: Field Studies (32805)
* Nancy B. Rybicki, U.S. Geological Survey, Reston, Virginia
- 1:45 p.m. Viability of Submersed Macrophyte Propagules in the Potomac River:
Laboratory Studies (32805)
* Dwilette G. McFarland, WES, Vicksburg, Mississippi
- 2:00 p.m. The Importance of Preemption in Competitive Interactions (32577)
* R. Michael "Mike" Smart, WES, Lewisville, Texas
- 2:15 p.m. Competitive Interactions of Native Plants with Nuisance Species
in Gunter'sville Reservoir (32736)
* Robert D. Doyle, WES, Lewisville, Texas
- 2:30 p.m. ADJOURN
- 3:00 p.m. Tour WES Facilities
- 5:00 p.m.

Thursday, 17 November 1994

- 8:30 a.m. General Session
- 12:00 Auditorium

Fish and Aquatic Plant Technology

Presiding: K. Jack Killgore, WES, Vicksburg, Mississippi

- 8:30 a.m. Overview of Fish and Aquatic Plant Technology
* K. Jack Killgore, WES, Vicksburg, Mississippi

- 8:45 a.m. Use of Multispectral Videography with GIS for Spatial Relationships Between Aquatic Vegetation and Fish (32944)
* Thomas "Tom" B. Hardy, Utah State University, Logan, Utah
- 9:00 a.m. Relationship Between Fish, Plants, and Turbidity (32944)
* Ray Drenner, Texas Christian University, Ft. Worth, Texas
- 9:15 a.m. Some Macrophyte, Invertebrate, and Fish Associations in a *Myriophyllum spicatum* L.: Will Increasing the "Edge" Change Anything? (32944)
* Thomas "Tom" D. Pellett, Wisconsin Department of Natural Resources, Madison, Wisconsin
- 9:30 a.m. Differences in Aquatic Plant Spatial Complexity and Its Effect on Foraging Largemouth Bass (32944)
* Eric Dibble, WES, Vicksburg, Mississippi
- 9:45 a.m. BREAK

Simulation Technology

John D. Madsen, WES, Lewisville, Texas

- 10:15 a.m. An Overview of Simulation Technology Development
* John D. Madsen, WES, Lewisville, Texas
- 10:30 a.m. Development of Plant Population Simulation Models (32440)
* R. Michael "Mike" Stewart, WES, Vicksburg, Mississippi
- 10:45 a.m. Biological Control Simulations: The AMUR Stocking Rate Model (32438)
* William "Will" A. Boyd, WES, Vicksburg, Mississippi
- 11:00 a.m. A Herbicide Dissipation and Effects Model (32439)
* R. Michael "Mike" Stewart, WES, Vicksburg, Mississippi
- 11:15 a.m. Digital Spatial Database Technology Support to Simulation Modeling (32506)
* M. Rose Kress, WES, Vicksburg, Mississippi
- 11:30 a.m. Development of an Aquatic Plant Management Strategy Planner (32954)
* John D. Madsen, WES, Lewisville, Texas
- 11:45 a.m. Report on Tuesday's USAE Division/District Working Session
* Wayne T. Jipsen, USAE District, Jacksonville, Florida
- 12:00 noon ADJOURN 29th Annual Meeting
- 1:00 p.m. **FY 1996 Civil Works R&D Program Review**
- 4:00 p.m. (Corps of Engineers Representatives Only) Main Conference Room

Poster and Demonstration Session

Posters:

Facilities for Biocontrol Research

- *Alfred "Al" F. Cofrancesco, WES, Vicksburg, Mississippi
- Edward Snoddy, Tennessee Valley Authority, Muscle Shoals, Alabama
- William "Bill" K. Oldland II, TVA;
- Michael "Mike" J. Grodowitz, WES

Biocontrol Agents of Aquatic and Wetland Plants

- *Biomangement Team, WES, Vicksburg, Mississippi

Competitive Effects of American Lotus on Eurasian Watermilfoil (32736)

- *Robert D. Doyle, WES, Lewisville, Texas

Ecological Assessment of Kirk Pond, Oregon

- *John D. Madsen, WES, Lewisville, Texas

Detection and Mapping of Submersed Vegetation Using Hydroacoustic, GPS, and GIS Technologies

- *Bruce M. Sabol, WES, Vicksburg, Mississippi

Lewisville Aquatic Ecosystem Research Facility (LAERF)

- *R. Michael "Mike" Smart, WES, Lewisville, Texas

Aquatic Plant Control Operations Support Center (APCOSC)

- *Wayne T. Jipsen, USAE District, Jacksonville, Florida

Environmental Protection Agency (EPA)

- *Fred Kerpel, USEPA, Atlanta, Georgia

Demonstrations:

Information/Expert Systems

- *Michael "Mike" J. Grodowitz, WES, Vicksburg, Mississippi

HERBICIDE Simulation Model (32439)

- *Joel A. McAllister, WES, Vicksburg, Mississippi

AMUR Stocking Rate Model (32438)

- *William "Will" A. Boyd, WES, Vicksburg, Mississippi

Aquatic Plant Management Strategy Planner Demonstration Shell (32954)

- *Joel A. McAllister, WES, Vicksburg, Mississippi

Aquatic Plant Bulletin Board System (APBBS)

- *Joel A. McAllister, WES, Vicksburg, Mississippi

Geographic Information System Technology to Support Aquatic Plant Control Programs (32506)

- *E. May Causey, WES, Vicksburg, Mississippi

Attendees

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Conversion Factors, Non-SI to SI Units of Measurement

Non-SI units of measurement used in this report can be converted to SI units as follows:

Multiply	By	To Obtain
acres	4,046.873	square meters
acre-feet	1,233.489	cubic meters
degrees (angle)	0.01745329	radians
feet	0.3048	meters
gallons (U.S. liquid)	3.785412	liters
inches	2.54	centimeters
miles (U.S. statute)	1.609347	kilometers
ounces (mass)	28.34952	grams
pound (mass)	0.4535924	kilograms
quarts (U.S. liquid)	0.9463529	liters
square feet	0.09290304	square meters
tons (mass) per acre	0.22	kilograms per square meter

Introduction

The Corps of Engineers (CE) Aquatic Plant Control Research Program (APCRP) requires that a meeting be held each year to provide for professional presentation of current research projects and to review current operations activities and problems. Subsequent to these presentations, the Civil Works Research and Development Program review is held. This program review is attended by representatives of the Civil Works and Research Development Directorates of the Headquarters, U.S. Army Corps of Engineers; the Program Manager, Environmental Resources Research and Assistance Programs (ERRAP); and representatives of the operations elements of various CE Division and District Offices.

The overall objective of this annual meeting is to thoroughly review the Corps aquatic plant control needs and establish priorities for the future research, such that identified needs are satisfied in a timely manner.

The technical findings of each research effort conducted under the APCRP are reported to the Manager, ERRAP, U.S. Army Engineer Waterways Experiment Station, each year in

the form of periodic progress reports and a final technical report. Each technical report is distributed widely in order to transfer technology to the technical community. Technology transfer to the field operations elements is effected through the conduct of demonstration projects in various District Office problem areas and through publication of Instruction Reports, Engineers Circulars, and Engineer Manuals. Periodically, results are presented through publication of an APCRP Information Exchange Bulletin, which is distributed to both the field units and the general community. Public-oriented brochures, videos, and speaking engagements are used to keep the general public informed.

The printed proceedings of the annual meetings are intended to provide all levels of Corps management with an annual summary to ensure that the research is being focused on the current nationwide operational needs.

The contents of this report include the presentations of the 29th Annual Meeting held in Vicksburg, MS, 14-17 November 1994.

Center for Aquatic Plant Research and Technology

by
*J. L. Decell*¹

For nearly 20 years, the scientists and staff of the U.S. Army Engineer Waterways Experiment Station (WES) Environmental Laboratory (EL), through the Aquatic Plant Control Research Program (APCRP), have been conducting research for development of technology for managing problem species of aquatic plants that interfere with the valued uses of the Nation's water bodies. This research includes development of technology for the environmentally compatible use of chemical, biological, mechanical, and integrated control methods. In addition, ecological studies, simulation/modeling capabilities and environmental studies determining the relationships of aquatic plants to fisheries, water quality, and the aquatic ecosystem generally, are conducted. These comprehensive studies, the results of the research and the effective transfer of technology, have caused the APCRP, the EL scientists, and the EL APCRP staff to be recognized by other Federal agencies, state agencies, and universities as the national leader in the development of aquatic plant technology and conduct of research. In nearly all technical areas of endeavor, the APCRP and the respective lead scientists are also recognized internationally.

Aquatic plants occur in nearly every water body in the United States. Eventually, as part of natural processes, these plants interfere not only with navigation, flood-control activities, and water supply, but with hydropower generation, recreation, and, at very high population levels, with the natural balance of the quality of the aquatic ecosystems, often shortening the life of one or more components. The APCRP scientists have lead the Nation in developing and providing technology and innovative, environmentally compatible applications of technology for managing these

problem plants. Since 1977, two of the four major aquatic plant problems have been reduced to nonproblem levels, nationally, as a direct result of studies by the APCRP scientists and staff.

The APCRP, through its accomplishments of the last 15-20 years, is recognized as the national and international leader in the field of aquatic plant research and technology development. The APCRP has served the needs of the Corps of Engineers (CE) extremely well, and through the CE structure, has served the CE Districts' respective state cost-sharing partners. With the exception of the U.S. Department of Agriculture, Agricultural Research Service (USDA-ARS), the Tennessee Valley Authority (TVA), and the U.S. Department of Interior Bureau of Reclamation (USDI-BR), other Federal agencies and the majority of universities are not aware of this expertise and do not routinely turn to the APCRP for this technology. While the USDA-ARS, TVA, and the USDI-BR continually cooperate with the APCRP on research, noncooperating Federal agencies are generally unaware of the capabilities and expertise of APCRP. As aquatic plant managers of the United States are forced to deal with the ever-strengthening environmental concerns for all aspects of water resources management, the expertise of the APCRP and related sciences will be called on even stronger, to identify, develop, and apply the appropriate technology commensurate with the problem, and to do so in a manner that protects or enhances environmental quality. The Corps APCRP must be in a position, organizationally, to aggressively achieve the final plateau of recognition and responsiveness to the Nation through the other Federal agencies.

¹ Director, ERRAP, EL, U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

In August 1993, the Corps established the Center for Aquatic Plant Research and Technology (CAPRT) at the WES. The CAPRT will provide a single point of contact for coordination/facilitation office at WES for nationwide aquatic plant research and resulting technology to: (a) Congress and their staffs, (b) Army Staff, (c) Headquarters, U.S. Army Corps of Engineers (HQUSACE), (d) USACE Divisions, Districts, and Field Operating Activities (FOA's), (e) Installation Commanders and other Department of Defense (DOD) users, (f) other Federal agencies, (g) State agencies, (h) academia, and (i) private industry and organizations. The CAPRT will address Federal-wide aquatic plant management and technology coordination needs for both civil and military programs.

The CAPRT will provide administrative and technological leadership, facilitation and coordination of all aquatic plant research, technology development, and technology transfer. Activities will include: (a) direct allotted Research and Development (R&D), (b) technology transfer, (c) technical assistance, (d) workshops and seminars, (e) technical guidance documents, (f) general information requests, and (g) coordination with special interest groups/organizations.

Subsequent to its establishment, Mr. J. L. Decell was appointed Director of the CAPRT in 1993, with the authority and responsibility to execute the CAPRT mission. Key to this responsibility will be coordination with other Federal agencies engaged in aquatic plant research/management activities.

In late 1994, key Federal executives, representing agencies involved in aquatic plant control, were invited to hold Charter Membership in the CAPRT and serve as members of an advisory board to guide the CAPRT through the formative years and into the future. The Advisory Board for Center Activities (ABCA) was formed and consists of:

Dr. Mary E. Carter
Area Director
South Atlantic Area
USDA Agricultural Research Service
Athens, GA

Dr. Ronald Ritchard
Senior Scientist
Tennessee Valley Authority
Muscle Shoals, AL

Mr. Donald Stubbs
Chief, Fungicide-Herbicide Branch
Registration Division
U.S. Environmental Protection Agency
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Dr. Stanley Ponce
Chief, Research and Laboratory
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Denver, CO

The first meeting of the ABCA will be held during this meeting. It is planned that the ABCA will meet no more than once each year to review activities of the Center and provide guidance to ensure that all agencies' needs are being addressed, and that the Center's functions are being efficiently provided, not only for the Federal agencies, but also for those state and local agencies managing aquatic plant problems throughout the Nation.

Part of the concept of the CAPRT was to facilitate action items of common interest to other Federal agencies. This can be especially useful when regulatory requirements are part of the action. To be well informed on the correct issues, in a timely manner, this type of coordination must be effected at the working level. The CAPRT was established with the ability to form working groups as needed. These will be subject-matter-related groups and will remain in existence as long as the need exists. The first such working group to be formed is the Aquatic Herbicide Working Group (AHWG), with the initial purpose of more effectively communicating with the U.S. Environmental Protection Agency (USEPA) regarding aquatic herbicide registration matters. Dr. Kurt Getsinger was selected to serve as Chairman of the AHWG. Members of the AHWG include working level representatives from the EPA, USDA-ARS, USDI-BR, and TVA.

The issues being addressed and coordinated by the AHWG are also of vital interest to the herbicide industry. For that reason, industry representatives were invited to a meeting

during which an invitation was extended for industry to form a Herbicide Industry Working Group (HIWG). This group is not an integral part of the CAPRT and functions outside of the CAPRT. Its interests are functionally linked to the AHWG by agreement that the Chairman, AHWG, will also serve as a member of the HIWG. In this manner, the two groups can maintain continuity, while allowing the CAPRT to maintain its necessary status as a Federal Center.

Future plans for continued development of the CAPRT include the formation of an

Aquatic Biocontrol Working Group (ABWG) to coordinate actions for the development and use of biological control agents for management of aquatic plants. In addition, initiatives will be taken to determine the proper and appropriate involvement of interest groups that may have input into the development of future management practices.

It is hoped that the members of the CAPRT can identify issues and objectives of common interest to all involved Federal agencies, and can subsequently progress together to meet our respective goals.

Annual Report - Aquatic Plant Control Operations Support Center

by
Wayne T. Jipsen¹

In October 1980, the Jacksonville District was designated by HQUSACE as the Aquatic Plant Control Operations Support Center (APCOSC) in recognition of the District's knowledge and expertise gained through the administration of the largest and most diverse aquatic plant management program in the Corps. The APCOSC personnel assist other Corps elements and other Federal and state agencies in the planning and operational phases of aquatic plant control (APC).

The specific duties of the APCOSC and relationships with other Corps APC programs as outlined in ER 1130-2-412 are:

- Provide operational guidance to Corps Districts in the planning phases of APC programs.
- Provide technical guidance to Corps Districts in the operational phases of APC programs.
- Provide operational expertise and/or personnel and/or equipment to respond to localized, short-term critical situations created by excessive growths of aquatic plants.
- Provide assistance to HQUSACE and Division offices for the training and certification of Corps application personnel.
- Assist WES in the field application and evaluation of newly developed control techniques or procedures.
- Provide assistance to HQUSACE in the development and administration of a comprehensive Corps-wide APC program.

The demand for and types of services performed by the Center vary from year to year, based on the type of problems encountered by Corps elements and other agencies. Four basic types of information are requested: planning, operations, research, and training. Planning assistance includes determinations of water body eligibility and allowable costs, computation for benefit-cost ratios, methods of data acquisition, and other factors that enter into the process of planning an APC program. Operations assistance involves most aspects of chemical, mechanical, biological, and integrated technology. The Center provides data, information, and recommendations relating to operational activities. Information on research activities is provided to requestors if available, or the requests are referred to WES. Training assistance includes providing materials for use in educational and training programs and presentation of the Pesticide Applicators Training Course and the Aquatic Plant Management Course by Center staff.

During fiscal year 1994 (FY94) the Center responded to 144 requests for assistance. Figure 1 indicates the types of information requested; Figure 2 provides a breakdown regarding source of information requests.

Center activities during FY94 included providing answers to numerous questions on the development of private contracts for control operations, new-start programs, and economic impacts. In addition, the Center provided crews and equipment to assist WES researchers with the Tricopyr studies on Lake Minnetonka. Center training activities included airboat training, biocontrol training, and a variety of presentations to user groups and professional societies.

¹ U.S. Army Engineer District, Jacksonville; Jacksonville, FL.

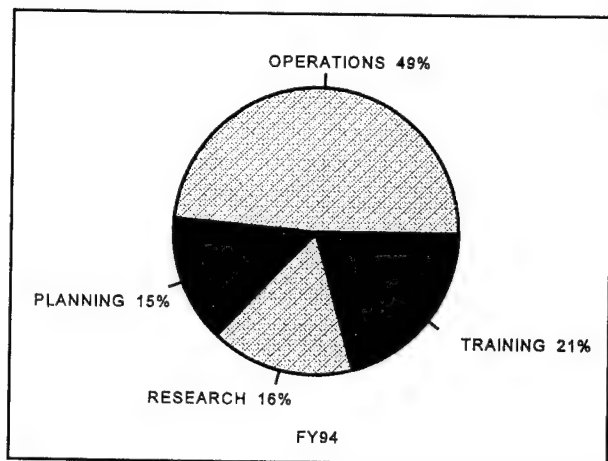


Figure 1. Types of assistance requested

A review of information requests for the past three years was undertaken to assist the Center staff in preparing for the future. This activity provided an insightful view of the overall program, as well as current and potential future concerns.

The three-year comparison of request types is provided in Figure 3. Figure 4 contains the comparison of the source of these requests. These comparisons document a number of phenomena. First, while the operations component of these requests remains steady at approximately 50 percent of all requests, FY94 saw a 33 percent increase in the number of planning assistance requests. In addition, training requests continue to increase.

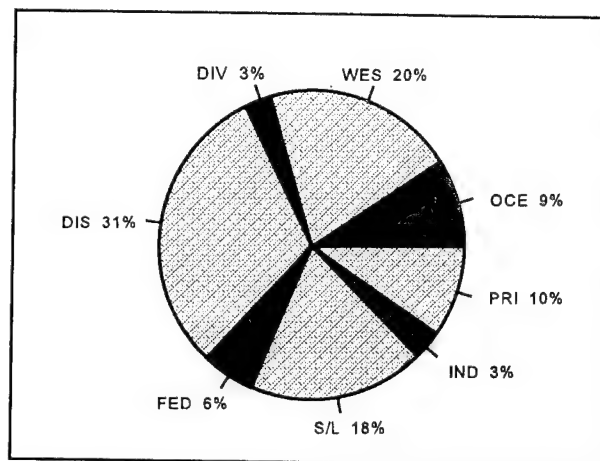


Figure 2. Sources of requests, FY94

Results of the on-going restructuring of the Corps Operations elements are evident in the comparison of sources of requests. The number of requests from Corps Division offices has decreased significantly while the number of requests from District offices has increased.

The increase in planning-related requests, along with the increase in requests from both District offices and State and local agencies can be attributed to an increased interest in program expansions and new program starts. These interests can be attributed to both the expansion of the range of exotic species and an increasing view of the APC program as an environmentally friendly program.

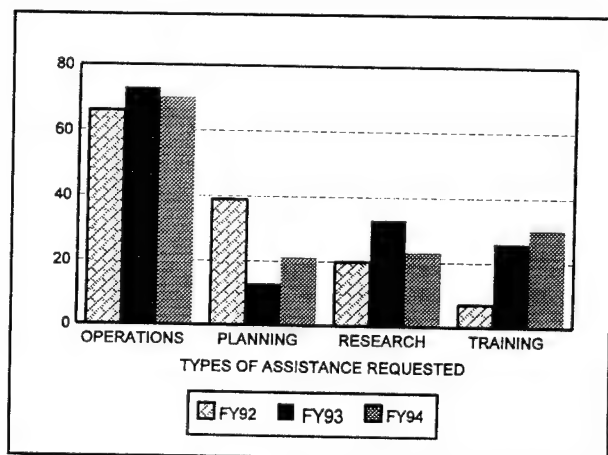


Figure 3. Three-year comparison of request types

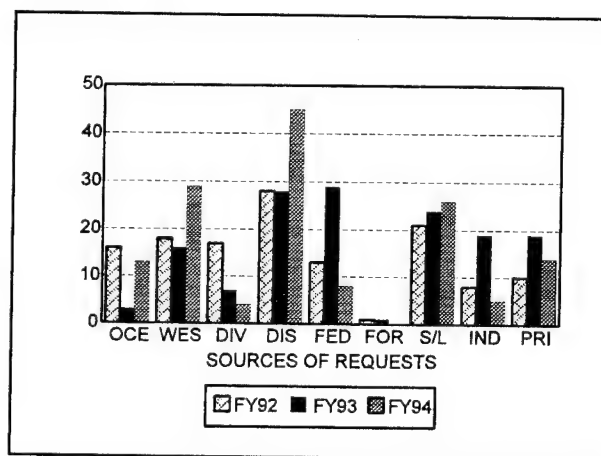


Figure 4. Three-year comparison of sources of requests

One of the main challenges of the Center as well as of all aquatic plant managers will be to provide an adequate level of training assistance to our partners and customers. Training continues to be one of the keys to

long-term control of exotic plants. This training needs to include a wide range of audiences in order to help slow the spread (especially intentional plantings) of the target species.

Use of Economic Information in the Evaluation of Aquatic Plant Control Programs: The Lake Guntersville Recreation Study

by
*Jim E. Henderson*¹

Decisions on aquatic plant control should be based on information on the preferences, perceptions, and values related to plant control and plant control alternatives. The Recreation Study component of the Guntersville Joint Agency Project (GJAP) is the most extensive study to date to analyze the public preferences and economic values of aquatic plant control for recreation (Thunberg 1991). Different user groups may hold differing and conflicting preferences for plant control that must be balanced and accommodated in planning for management of non-native vegetation (Henderson 1991). Conflicts derive from the differences between the desire of anglers for as much fishery habitat (aquatic vegetation) as possible and the desire by nonanglers for minimal vegetation to avoid interfering with water skiing, swimming, and pleasure boating (Henderson Draft).

Decisions about aquatic plant control programs must balance the needs and preferences of anglers with those of boaters and other non-angler recreational interests. The results of the Lake Guntersville Recreation Study will be used to show how the perception, preference, and valuation information is used to evaluate the impact of changes in aquatic plants on angler and nonangler users.

Results of the Study

As reported in previous meetings (Henderson 1994), surveys were used to elicit preference and valuation data from individuals. Three separate surveys were conducted to obtain (1) baseline recreation use and preference information; (2) recreation use and economic

value for the different alternatives; and (3) economic impact information (Bergstrom et al. 1994).

Baseline recreation use and preferences

Baseline recreation use and preference, importance, and satisfaction data were obtained by asking lake users to describe their experience and express their opinions. Anglers and nonanglers were asked the same questions. Respondents were asked to identify the recreation activities participated in and the amount of time spent at the lake.

Baseline recreation use and aquatic plant perceptual data were collected through face-to-face (onsite) interviews and mailed surveys. To use the lake, recreators either use a public access point, such as a park or boat ramp, or access is obtained for residential users by their lakefront property or common access lot (for backlots that do not front the shoreline). A face-to-face survey at public access points was used at marinas, boat ramps, and parks. Because of the difficulty of finding residents at home for a survey, a mailed survey was used for residential users (Henderson 1994).

Recreation use. Based on the onsite and residential surveys, a total of 3.1 million annual visits were estimated broken out as 2,844,718 onsite and 208,221 residential visits. In comparing anglers to nonanglers for total visits there were 1,066,175 angling visits and 1,975,917 nonangling visits, for a 35:65 percent angler:nonangler breakdown. The user group breakdown for the 2.8 million onsite

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

visits is further disaggregated as shown in Table 1 (Bergstrom et al. 1994).

Preferences. To obtain perceptions on the impact of aquatic plants on recreation, respondents were asked to characterize the impact as "A Help," "Doesn't Affect," "Bothersome Sometimes," or "Bothersome Most of the Time." To obtain preferences for the amount of plants, the response categories were "As Much as Possible," "More than Presently Exists," "Same as Presently Exists," "Less than Present," and "None." Residential respondents were asked to describe the aquatic plants near their home as "Not Noticeable," "Slightly Noticeable," "Moderate," or "Heavy/Severe."

Perceptions of aquatic plants showed differences between user groups (Table 2) (Bergstrom et al. 1994). Considering the impact of aquatic plants on recreation, for the onsite visitors, boaters and shore-based recreators clearly expressed that the plants had no effect (Table 2). Approximately half (47.1 percent) of the boat anglers view the

plants as a help to their recreation experience. The bank anglers are more widely split in evaluation, with only a fifth (20.1 percent) of the bank anglers seeing the plants as a help, half (54.2 percent) saying no effect, and a fourth of the bank anglers (25.7 percent) saying the plants are bothersome at least part of the time.

Perceptions on recreation impact by the residential users (Table 3) showed that residential users experienced greater negative impact by the plants (Bergstrom et al. 1994). With the exception of boat anglers (28.4 percent), each group responded as bothersome at least part of the time, bank anglers (74.1 percent), boaters (70.7 percent), and no main activity (53.7 percent).

Respondents that indicated plants had an effect on their recreation were asked preferences for amounts of plant coverage (Table 4) (Bergstrom et al. 1994). Presenting the onsite results, an increase in plant coverage (more plants or as much as possible) (i.e., reduction

Table 1
User Group Annual Recreation Visits - Onsite Survey

User Group	Total Onsite Recreational Visits	Confidence Interval (90%)	Percent of Total Visits
All users	2,844,718.14	29.63%	100
Angler/Boater	645,224.32	16.74%	22
Angler/Nonboater	331,284.15	63.92%	12
Nonangler/Boater	271,807.07	25.92%	10
Nonangler/Nonboater	1,596,402.60	50.97%	56

Table 2
Impact of Aquatic Plants on Recreation (percent), Onsite Survey

Main Activity	Helps	Does Not Affect Activity	Bothersome Sometimes	Bothersome Most of the Time
Bank angling	20.1	54.2	15.3	10.4
Boat angling	47.1	36.0	9.9	7.1
Boating	4.1	74.4	11.9	9.6
Shore-based recreation	7.2	86.4	4.0	2.4
All visitors	26.9	58.1	8.7	6.2

Note: Chi-square = 451.1; $p < 0.0001$.

Table 3
Impact of Aquatic Plants on Recreation (percent), Residential Users

Main Activity	A Help	Does Not Affect	Bothers Sometimes	Bothersome Most of the Time
Bank angling	12.9	12.9	41.9	32.2
Boat angling	50.5	20.8	18.6	9.8
Boating	2.6	26.5	51.3	19.4
No main activity or other	5.5	40.6	34.4	19.3
All residents	16.0	29.4	36.3	18.1

Note: Chi-square: $p < 0.001$.

Table 4
Preferences for Amounts of Aquatic Plant Coverage, Onsite Survey (percent)¹

Main Activity	As Much as Possible	More Than Presently Exists	Same as Presently Exists	Less Than Present	None at All
Bank angling	11.1	17.3	35.8	24.7	11.1
Boat angling	28.9	33.1	20.3	13.6	4.1
Boating	0.0	1.4	29.6	42.3	26.8
Shore-based recreation	20.9	9.1	22.7	36.4	10.9
All visitors	23.9	26.0	22.8	19.8	7.5

¹ Only those visitors who said aquatic plants have some effect on their recreational activity responded to this question.
Note: Chi-square = 174.6; $p < 0.0001$.

in total control) is favored by 62 percent of the boat anglers. The bank anglers are fairly evenly divided with 28.4 percent preferring more plants (or as much as possible), 35.8 percent satisfied with existing conditions, and 35.8 percent wanting less or none at all. Sixty-nine percent of the boaters prefer less plants or none at all, with another 29.6 percent preferring present conditions. About half (47.3 percent) of the shore-based recreators preferred less plants than current conditions.

Perceptions were obtained on the importance and satisfaction of a range of recreation and resource management attributes such as opportunity to catch a lot of fish, open water areas, and availability of camping, swimming, picnic, and camping facilities. The importance and satisfaction evaluations provided extensive amounts of information on a range of resource management and recreation characteristics that assists in evaluation of impacts of angler/nonangler groups. A portion of

these results will be presented in the following section where the impact of different aquatic plant alternatives on user groups are evaluated.

Recreation use and economic value of plant control alternatives

The economic valuation of plant alternatives was determined using a contingent valuation method (CVM) survey. The CVM survey was administered as a mailed survey, using the residential survey sample and names collected as part of the onsite survey (Henderson 1994). The CVM approach presents alternative scenarios, in this case alternative plant control alternatives. The scenarios were depicted graphically and verbally in enough detail so respondents can evaluate what their response would be to the alternative plant conditions. That is, the response or behavior is contingent on the scenario conditions.

Five alternative plant control scenarios were presented (Table 5) covering the range from no control of plants (minimal control) estimated to be about 34,000 acres of the lake (50 percent of the lake acreage) to complete eradication of the plants (Alternative D). The three intermediate alternatives represent the vegetation during past years (Henderson 1994).

Table 5
Lake Gunterville Aquatic Plant Control Alternatives

Alternative	Plant Coverage, acres	Year
Minimal control	34,000	
Alternative A	20,000	1988
Alternative B	14,000	1989
Alternative C	8,000	1990
Alternative D	Near "0"	

Analysis of the CVM survey provided decisionmakers at all levels with data to compare effects on users of different levels of plant control. Differences in economic values and use patterns for anglers and nonanglers further enable comparison of differential impacts on user groups. Respondents were classified as anglers and nonanglers and separate valuation equations were estimated for the two groups. Estimates of recreation visits were developed by logistic regression equations. Potential variables included in the regression equations were the bid amount, management alternative, demographic variables (income, age, sex, lake residence status, family size), availability of recreation substitutes, and an

index of the amount of plants preferred by the individual.

Annual recreation use for the five alternatives was calculated from responses to the CVM scenarios (Table 6) (Bergstrom et al. 1994). Total annual visits were estimated from responses to the five CVM scenarios. Measurement of recreation visits is important because it infers desirability and acceptability of alternative aquatic plant conditions for recreation. The aggregate or total visits is used to calculate the aggregate economic value of aquatic plant control, described below.

Recreation use (number of visits) was highest for Alternatives A, with Alternative B following closely with only about a 3 percent difference (Table 6). The user group composition of visitation is different, with Alternative A having a majority of angler visits while Alternative B has a higher proportion of nonangler visits.

In the valuation models estimated for angler and nonangler groups, the regression equations estimated the probability that respondents would say "yes" to the bid amounts. The probability functions were then integrated over the bid variable values. The resulting angler and nonangler mean annual economic values for each alternative are shown in Table 7. The variables that were significant and included in the models were: bid amounts, income of respondent, residence status (whether the respondent lived on the lake), and the substitute variable (the availability of substitute water recreation sites for the respondents), significant for anglers only.

Table 6
Estimated Visits by User Groups and Management Alternatives

User Group	Minimum Control Alternative	Management Alternative A	Management Alternative B	Management Alternative C	Management Alternative D
Angler	1,644,440	1,847,554	1,429,608	976,508	476,536
Nonangler	855,640	1,434,785	1,765,458	1,868,209	1,750,512
Total visits	2,500,080	3,282,339	3,195,066	2,844,717	2,227,048

Table 7
Mean Annual Economic Values per Visitor for Angling and Nonangling Groups (1990 dollars)

Control Alternative	Anglers	Nonanglers
Minimum control	\$790.47	\$-334.80
Alternative A	778.46	424.83
Alternative B	503.77	918.84
Alternative C	9.43	1,050.37
Alternative D	-639.87	999.94

The anglers had the highest mean annual value for the Minimum Control Alternative while Alternative C is highest valued for the nonanglers (Table 7). Alternatives A and B provide significant economic values for both user groups. The negative values in Table 7 indicate recreation users would have to be compensated to use the lake.

The aggregate annual economic value of each alternative for all user groups (Table 8) was calculated based on the number of visits (Table 6) and user groups mean annual economic values (Table 7). The aggregate economic value is the total economic value of recreation for all user groups, anglers and nonanglers. Alternative B has the highest annual economic value with an aggregate total of \$122 million, followed by Alternative C and Alternative A (Table 8). Two things of note about this result. First, the alternative with highest aggregate economic value (Alterna-

Table 8
Aggregate Annual Net Economic Value for Management Alternatives

Alternative	Aggregate Annual Economic Value
Minimum control	\$21,591,799.83
Alternative A	93,979,867.14
Alternative B	122,795,210.20
Alternative C	101,799,931.70
Alternative D	52,721,076.02

tive B) is not the highest valued alternative for either the angler or the nonangler user groups (Table 7). The second consideration is in comparing the alternative aggregate values to the number of estimated visits (Table 6) for each alternative. Alternative A has a slightly higher number of visits (Table 6), but Alternative B has the highest aggregate economic values (Table 7). The effect is likely the result of differences in the mean annual economic values of the user groups (Table 7) and the differences in visits by the groups under each alternative (Table 6). Alternative B has a higher number of nonangler annual visits (compared to Alternative A) which are higher valued than the anglers, resulting in a higher aggregate economic value.

Economic impact to the local economy

The analysis of economic impacts to the local economy was based on the expenditure survey. Lodging, food, gas, supplies, and fishing tackle are examples of recreation expenditures for goods and services of interest to the local economy (Henderson Draft).

To develop estimates of economic impact, an expenditure survey was given to onsite respondents, to be completed at home and mailed back. Expenditure surveys were mailed to residential respondents. Respondents were asked to summarize their recreation expenditures for trips to Lake Guntersville and for annual recreation expenditures, e.g., licenses, boats, motors, fishing tackle, summarized at Table 9. Nonangler trip expenditures are approximately 18 percent more than anglers (1990 dollars).

The expenditure summaries were used in an economic input-output model (IMPLAN) that estimates the economic impact on specific economic sectors important to recreation (Palmer and Siverts 1985, Probst 1988). IMPLAN allocates expenditures to the appropriate economic sector. When an angler buys, for example, fishing tackle at a store at Lake Guntersville, those expenditures represent the direct or "first round" effects of recreation spending. Increases in demand for fishing

Table 9
Expenditure Summary by Expenditure Categories

User Type	Lodging	Food	Transportation	Activities	Misc.	Total
Angler	\$22.88	\$24.23	\$32.12	\$4.17	\$4.98	\$88.38
Nonangler	4.28	47.09	40.21	2.82	13.92	108.32

tackle, caused by increased recreation, cause increases in orders for tackle. Tackle manufacturers purchase the raw materials and labor necessary to meet the new orders for tackle. The increases in purchase of raw materials, labor, and other items by the tackle manufacturers are known as indirect effects. The increased employment and income resulting from additional tackle production puts more income in the hands of tackle manufacturers, suppliers, and their employees; this income is then spent on additional goods and services. This spending resulting from increased income is known as the induced effects (Bergstrom et al. 1994).

The IMPLAN model accounts for and allocates the direct, indirect, and induced spending effects to the appropriate economic sectors.

IMPLAN estimates economic impacts (Table 10) in terms of increases in total gross output or production, e.g., food service, tackle, boats; income; and employment associated with each of the alternatives. These values represent economic impact as increases resulting from recreation expenditures associated with each of the alternatives.

Economic impacts are summarized in Table 10, showing increases in total gross output, income, and employment as a result of user-group spending associated with the five alternatives. These economic impacts were estimated for the two-county area where Lake Guntersville is located. Comparing alternatives, the pattern is similar for all three measures of economic impact. Alternative A produces the greatest increase in gross production, income,

Table 10
Economic Impacts Due to Recreational Spending

User Type	Minimum Control Alternative	Management Alternative A	Management Alternative B	Management Alternative C	Management Alternative D
Total Annual Gross Output Due to Recreational Spending at Lake Guntersville, \$ million (1990 dollars)					
Angler	155.19	174.36	134.92	92.16	44.69
Nonangler	86.36	144.81	178.19	188.55	176.68
Total	241.55	319.17	313.11	280.71	221.37
Total Annual Income Due to Recreational Spending at Lake Guntersville, \$ million (1990 dollars)					
Angler	87.78	98.63	76.31	52.12	25.43
Nonangler	48.70	81.66	100.48	106.33	99.63
Total	136.48	180.29	176.79	158.45	125.06
Total Employment Due to Recreational Spending at Lake Guntersville, Total Number of Jobs					
Angler	4,608	5,177	4,066	2,736	1,335
Nonangler	2,681	4,496	5,532	5,854	5,485
Total	7,289	9,673	9,598	8,590	6,820

and employment. However, Alternative B is only about a percentage point below Alternative A on all three measures. This result is likely due to the small (3 percent) difference in total visits between the two alternatives.

Considering that Alternative A represents 30 percent coverage of the lake versus 20 percent for Alternative B, the difference in economic impacts resulting from this large (10 percent) change in plant coverage is minimal. Looking at this from the standpoint of the marina operator or the bait seller, a large difference in plant coverage (at the 20 to 30 percent level) does not translate into a significant difference in the amount of economic activity.

Applications of the Study Findings

Decisions on changes in aquatic plant control programs should take into account the preferences, values, and behavior responses (visits) of angler and nonangler groups. The economic value and alternative evaluations presented above provide insight to understanding angler and nonangler responses to changes in aquatic plant control and can be used to evaluate public acceptance of different control alternatives.

In evaluating the public response to changes in aquatic plant control, there are two questions to be considered. The first question is: How will recreation behavior likely change in response to altered plant conditions? Specifically, what will be the changes in visitation and economic value resulting from a change. The second question for consideration is: What are the angler and nonangler user group perceptions related to the change in amount

of vegetation, and how will the recreation behavior likely change in response to altered plant conditions?

In evaluating changes in aquatic plant control, three situations are likely: reduction in total control resulting in an increase in plants; increase in plant control to increase open water areas; and a change in control around shorelines (Henderson Draft). The positive and negative impacts of these plant changes on user groups are summarized in Table 11 and discussed with relation to study findings presented above.

Reduce total control

A reduction in the total amount of plant control results in an increased amount of aquatic plants. This situation can be caused from a reduction in control efforts by the reservoir operations agency or from changes in natural resource conditions that alter growing conditions affecting plant growth, e.g., drought (Henderson 1991). This point is readily understood by looking at the reductions in plant coverage reflected in the years 1988-1990 (Table 5). These significant reductions were primarily attributed to a series of high water years which reduced the growing conditions, and not entirely the result of increased control efforts.

Evaluating a change in total plant control starts with examination of perceptions of the impact of existing plant conditions (Table 2). Boaters and shore-based recreators clearly expressed that the plants had no effect, 74.4 and 86.4 percent, respectively. Approximately half (47.1 percent) of the boat anglers view the plants as a help to their recreation experience. The bank anglers are more widely split

Table 11
Summary of Impacts of Changes in Aquatic Plant Control

Change in Plant Control	Positive Impact	Negative Impact
Reduce total control	Boat anglers	Boaters
Increase open water areas	Boaters	Shore-based recreators
Change in shoreline control	Bank anglers	Boat anglers

in evaluation, with only a fifth (20.1 percent) seeing the plants as a help, half (54.2 percent) saying no effect, and a fourth of the bank anglers (25.7 percent) saying the plants are both-ersome at least part of the time.

Looking at preferences for amounts of plant coverage (Table 4) (asked of respondents indicating the plants had an effect on their recreation), an increase in plant coverage, more plants or as much as possible (i.e., reduction in total control) is favored by 62 percent of the boat anglers. The bank anglers were fairly evenly divided with 28.4 percent preferring more plants (or as much as possible), 35.8 percent satisfied with existing conditions, and 35.8 percent wanting less or none at all. The boaters would be negatively impacted by an increase in plants (Table 4). Sixty-nine percent of the boaters preferred less plants or none at all, with another 29.6 percent preferring present conditions. About half (47.3 percent) of the shore-based recreators preferred less plants than current conditions.

To summarize, a change in plant control that increases the amount of plant coverage would be favored by boat anglers and would negatively impact boaters and shore-based recreators. Looking at the relative amount of visits affected, boaters represent 10 percent of the visits, boat anglers 22 percent, and shore-based recreators 56 percent (Table 1). The point is that the number of total visits (or impact on visitation) by a group should be considered in using the preference information to make decisions. While the strength of prefer-

ences may be high or extreme, the total amount of visitation affected should also be considered. Sixty-nine percent of the boat anglers prefer more plants, and that preference represents only 10 percent of the total number of visits (Tables 1, 4).

Based on the estimate of visits under each alternative (Table 6), with increased plants the number of visits would increase to a maximum under Alternatives A and B, with Alternative B having more nonangling visits and Alternative A more angling visits. With increasing amounts of plants, annual economic values would increase for the angling group and decrease for the nonangling group.

Increasing open water areas

Many lake managers are under pressure to increase open water areas for use by pleasure boaters and more recently by jet skiers and other watercraft requiring wide expanses of water to avoid congestion and for safety. Increasing open water areas positively affects the boater group by providing more recreation areas and negatively affects boat anglers by reducing fishery habitat.

Looking at the satisfaction and importance analyses, satisfaction levels for boating and angling attributes (Table 12) show greater satisfaction levels for boaters. Characteristics affecting the boating group (open water areas, water skiing opportunities) are relatively high at mean values of 4.13 and 4.08, respectively (1-5 scale). Satisfaction levels for characteristics

Table 12
Satisfaction Levels (Means) for Boating and Boat Fishing Characteristics Onsite Survey

Characteristic	Bank Fishing	Boat Fishing	Boating	Shore-based
Open water areas	3.85	3.82	4.13	4.02
Water skiing opportunities	3.56	3.95	4.08	3.92
Opportunity to catch a lot of fish	3.58	3.49	3.80	3.67
Opportunity to catch large fish	3.52	3.50	3.72	3.68

Note: Means are based on a 5-point Likert scale, 1 = Terrible, 5 = Delighted.

important to anglers (opportunity to catch a large number of fish and opportunity to catch large sized fish) are lower. Mean satisfaction values for boat angler attributes are 3.49 for catching large numbers of fish and 3.50 for catching large sized fish (Henderson Draft). As discussed with the perception and preference results, satisfaction levels for characteristics affecting boaters are higher than satisfaction levels for the boat anglers (Table 5). This is indicative that boating conditions are highly satisfactory and that it may be difficult to increase that satisfaction level significantly. In other words, it may not be possible to achieve measurably higher levels of satisfaction for the boaters.

Satisfaction levels for the boat anglers may be further diminished if further reductions in plants occur, losing fishery habitat. If increased open areas of water are requested it may be possible to redistribute boat angling use to another part of the lake by lake use restrictions or by constructing new access points in areas of extensive plants.

Change in plant control around shorelines

In aquatic plant control programs, a major emphasis is placed on maintaining access to the lake by cutting boat lanes to residential boathouses, boat docks, and residential shorelines. Additionally, public swimming beaches and other areas of high use are kept free of plants.

Looking at residential responses to changes in shoreline control, residential respondents were asked to describe the aquatic plant coverage near their homes (Table 13). Those anglers fishing from piers, boathouses, and residential shoreline property (bank anglers) are the group most affected by residential shoreline conditions. This group reported plant coverage as heavier than the other user groups. Over half of the resident bank anglers (51.5 percent) described plant coverage as moderate or heavy/severe. The bank angler percentages for the moderate or heavy/severe categories were higher (51.5 percent) than the boat anglers (32.6 percent) or the boater group (36 percent). Additionally, perceptions of impact on recreation (Table 3) for bank anglers show 74.1 percent of residential respondents perceive the plants as bothersome at least part of the time.

Residential respondents were asked the importance of plant control attributes around residential areas. Overall, residential groups show strong support of control efforts around the residential area (Table 14). A public concern has been the use of sprayed pesticides in the lake (Tennessee Valley Authority 1993), but there are high levels of support for use of spray pesticides. When asked statements about attributes related to control around boathouses and control to maintain access for residential areas, means for all user groups were in the Strongly Agree-Agree (1-2) range (Table 14).

Given perceptions of already high level of plants by the residential bank anglers, increased

Table 13
Aquatic Plant Coverage Near Residential Homes (percent)

Main Activity	Not Noticeable	Slightly Noticeable	Moderate	Heavy/Severe
Bank angling	16.1	32.2	32.2	19.3
Boat angling	42.3	25.0	23.9	8.7
Boating	23.6	39.4	22.8	14.0
No main activity or other	22.9	32.4	29.3	15.2
All permanent residents	27.1	32.7	26.4	13.7

Note: Chi-square: $p < 0.03$.

Table 14
Importance Means for Aquatic Plant Control Attributes at Residential Areas

Attribute	Bank Fishing	Boat Fishing	Boating	No Main Activity	Overall
Like clear shoreline for access	1.57	1.66	1.25	1.55	1.49
Support spraying aquatic plants near my boathouse	1.87	1.92	1.36	1.66	1.66
Aquatic plants decrease value of my property	1.87	2.04	1.50	1.72	1.74
Prefer clear boat lanes leading from my boathouse	1.55	1.65	1.32	1.46	1.47
Aquatic plant control necessary for management of residential access areas	1.71	1.74	1.35	1.57	1.56

Note: Means are based on a 5-point Likert scale, 1 = Strongly agree, 5 = Strongly disagree. Means significantly different at 0.01 level.

plant control around the shorelines should be welcomed by the bank anglers. As discussed, residential groups show strong support of control efforts around the residential areas, including use of spray pesticides.

Agreement on the statement about effects of aquatic plants on residential land values is supported by another GJAP study (Driscoll et al. in press). A hedonic analysis was used in a land values study to estimate the effect of aquatic plants on land values. Hedonic analyses determine the contribution of different attributes or characteristics to the total value. In the land values study, the hedonic analysis determined the influence of the presence and extent of aquatic plant coverage on residential land values. The analysis showed that complete aquatic plant control, that is, eradication of plants in front of lakefront homes (not backlots or undeveloped lots) at Lake Guntersville, increased residential values by \$12 million annually.

Summary and Recommendations

The highest level of economic value for recreation, \$122 million annually, is achieved at 20 percent plant coverage of the lake. This amount of plant coverage provides highest economic value when angling activity can be at high levels and nonangling recreation is

also high, not hindered by excessive plant growth. This level of plant coverage strikes an acceptable balance between angling and nonangling recreation.

The Lake Guntersville study of recreation preferences and valuation is the most extensive valuation of aquatic plant control (Thunberg 1991). The results presented herein are representative of angler and nonangler use at large multiple-purpose reservoirs constructed and operated for navigation, flood control, and other nationally important purposes.

Future plant control valuation studies should focus on two types of studies. A valuation study should be implemented at other large reservoirs to determine how similar preference and values are among the large reservoirs that provide the majority of the Nation's water-based recreation opportunities. It may prove insightful to evaluate a reservoir where the sportfishing interests and industry are less developed or prominent than at Lake Guntersville (Tennessee Valley Authority 1993).

The second study type should evaluate smaller resource systems or reservoirs. State-managed reservoirs or power company lakes should be considered. Smaller systems may be easier to control plant coverage on, but the likelihood of conflict between angling and nonangling use may be increased because of the smaller size resource.

The Lake Guntersville study evaluated alternatives, depicted for respondents as six different recreation environments. Numerous comments by respondents, resource managers, and recreation specialists noted that the total amount of plant coverage may not be as important as the distribution of the plants. That is, where the recreator encounters the plants may be more important for preference and valuation than the total plant acreage of an alternative. The Lake Guntersville study asked questions on the level of satisfaction and importance of total amount and distribution of plants. Future studies should address the distribution of plants in a more extensive and explicit manner. Preferences for types of lake areas, e.g., around boat ramps, where the plants should be controlled, and the areas where plants are acceptable or desired should be elicited in a detailed manner.

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Triploid Grass Carp Growth, Mortality, and Population Size in the Santee Cooper Reservoirs, South Carolina - 1994

by

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Introduction

The Santee Cooper reservoirs cover 70,000 hectares in South Carolina and consist of Lakes Marion and Moultrie and the diversion canal connecting the two reservoirs. Hydrilla (*Hydrilla verticillata*) became established in upper Lake Marion in the early 1980's (Inabinet 1985) and spread throughout the entire system colonizing approximately 22,000 hectares in 1994. Control by herbicides alone was not feasible and stocking of triploid grass carp (*Centropharyngodon idella*) as a biocontrol agent began in 1989. From 1989 through 1992, 100,000 triploid grass carp were stocked yearly; 50,000 and 152,500 fish were stocked in 1993 and 1994, respectively. The estimated stocking density is 5 fish/ hectare.

Triploid grass carp are sterile, have dietary preferences similar to diploid grass carp, and have potential for low-cost, long-term control of nuisance aquatic vegetation (Sutton 1985; Allen and Wattendorf 1987; Wattendorf and Anderson 1984). However, proper use of this fish requires that important population attributes such as population size (and hence density), growth, and total mortality be accurately estimated for use in the AMUR/STOCK model (Boyd and Stewart 1994).

Methods to collect and age triploid grass carp were developed that allow monitoring in large water bodies (Kirk, Morrow, and Killgore 1994). This article summarizes collection techniques, aging techniques, growth rates, mortality rates, and population estimates of triploid grass carp in the Santee Cooper reservoirs.

Methods

Initial collection attempts using conventional fisheries gears, such as electroshocking and gill nets, failed. Subsequently, the South Carolina Department of Natural Resources (DNR) assisted WES by recruiting skilled bowfishermen and managing collection efforts. This technique involved using a fishing boat configured with lights in which bowfishermen stood on the bow and collected grass carp at night (Kirk et al. 1993).

Length-to-weight relationships, determination of age using hard structures, and backcalculation of length at earlier ages were used in estimating population attributes. Length-to-weight relationships were used to estimate condition of fish and weight at backcalculated lengths. Scales, otoliths, and spines were examined for annual marks or annuli (Jearld 1983). Lengths at an earlier age were backcalculated and weights at this previous age were estimated using a length-to-weight relationship specific to the Santee Cooper reservoirs. For additional information on aging structures (lapillar otoliths and scales), age distributions, and growth see Kirk et al. (1993) and Kirk, Morrow, and Killgore (1994).

We used a catch curve (Ricker 1975) to estimate total mortality. The number of fish by age class (adjusted for the number of fish stocked) was plotted and the descending or decreasing arm of the plot was used for mortality calculations. The natural log of numbers of fish collected (dependent variable) was regressed against age classes (independent variable). The slope of the regression was the

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instantaneous rate of total mortality (Z). The annual rate of survival was the antilog of Z.

The population of triploid grass carp in the Santee Cooper reservoirs was estimated using the relationship (Ricker 1975):

$$N_t = N_0 e^{-zt}$$

Precise counts of the number of fish stocked (N_0), the instantaneous rate of total mortality (z), and the number of years since stocking (t) were used in calculations. Using a spreadsheet and this relationship, population size and biomass by year class were estimated.

Results

Collection of triploid grass carp using skilled bowfishermen proved to be an efficient, cost-effective method. During 1994, approximately 160 fish were collected from late winter through early fall; collection success varied from 0 to 28 fish per evening depending on weather conditions. More triploid grass carp were collected on calm nights with clear water than during windy weather.

The length-to-weight relationship was $\text{Weight} = 0.0000032\text{Length}^{3.25}$ and this relationship differed little from previous years (Kirk, Morrow, and Killgore 1994). Age-specific weights ranged from 0.38 kg to 15.2 kg for ages 1 through 6, respectively (Table 1).

These weights show a linear increase in weight of approximately 3 kg/year (Figure 1). Growth in length (Table 1) was rapid during the first three years (150 to 200 mm/year) but decreased to 60 to 70 mm yearly during ages 4 through 6.

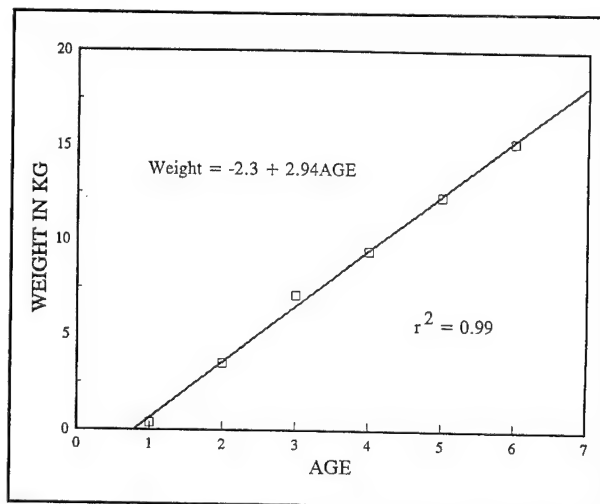


Figure 1. Growth of triploid grass carp stocked in the Santee Cooper reservoirs, South Carolina

The instantaneous rate of total mortality was estimated at -0.246 (Figure 2) which converts to an approximate annual rate of survival (S) of 80 percent. We estimated approximately 135,000, 39,000, 61,000, 48,000, 37,000 and 29,000 fish surviving in August 1994 for year age classes 1 through 6, respectively, and a total biomass of approximately 2,000,000 kg (Table 1).

Table 1
Population Attributes and Size at Age of Triploid Grass Carp in the Santee Cooper Reservoirs as of August 1994

Population Attributes			Age-Specific Attributes	
Age	Number Surviving	Biomass (kg)	Length (mm)	Weight (kg)
1	134,850	51,377	322	0.38
2	39,096	138,439	639	3.5
3	61,140	436,785	793	7.1
4	47,806	447,950	862	9.4
5	37,381	459,378	937	12.3
6	29,229	446,681	1,002	15.2

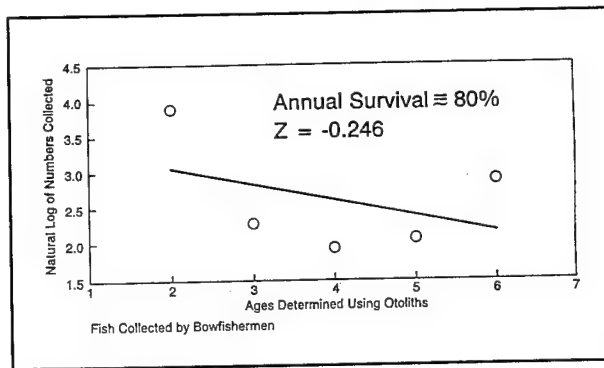


Figure 2. Catch curve used to estimate mortality of triploid grass carp stocked in the Santee Cooper reservoirs, South Carolina

Discussion

Accurate estimates of mortality, specifically the instantaneous rate of total mortality (Z) (Ricker 1975), are critical to proper use of the AMUR/STOCK model. Figure 3 illustrates the importance of accurate mortality estimates. As an example, an error of 20 percent in estimated mortality can cause a 300 percent error in the estimated population size after 4 years. Consequently, estimates of mortality need to be based upon correctly applied methodologies. Our mortality estimates were affected by higher than expected survival of the initial 1989 stocking (Figure 2). This trend suggests that annual survival over time may be less than 80 percent and populations should be

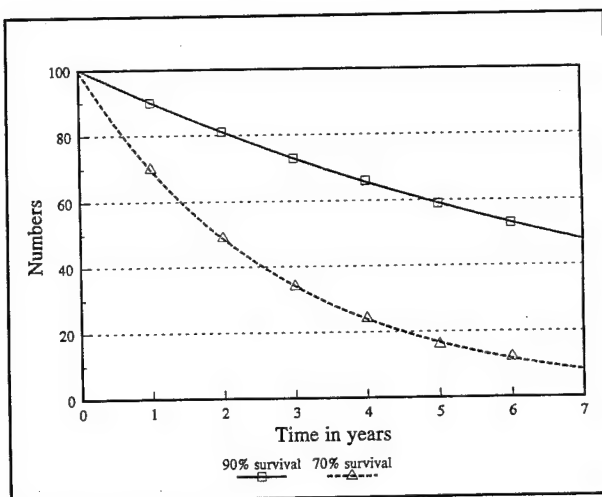


Figure 3. Decline of a cohort of 100 fish over 6 years with annual survival rate(s) of 90 and 70 percent

monitored yearly to properly support stocking strategies.

Earlier efforts (Kirk, Morrow, and Killgore 1994) using scales alone to estimate age distributions were improved by using otoliths. Scales from fish became difficult to read after age 4. We improved our estimates by using sectioned lapillar otoliths to determine age and then using scales of known age for backcalculations.

Growth in weight was estimated at approximately 3 kg/year. This rate was linear ($r^2 = 0.99$) (Figure 1) which may indicate that hydrilla, a preferred food, is not limiting. We speculate that growth may decline if hydrilla abundance decreases.

This work unit is ending in FY95. During this seven-year study we have monitored the movement of triploid grass carp in relation to food sources (Chappelear et al. 1991), studied water quality associated with beds of hydrilla (Kartalia, Foltz, and Killgore 1992), and surveyed native fish populations in relation to aquatic vegetation. We developed novel techniques to collect adequate numbers of grass carp for population monitoring and developed aging techniques (Kirk et al. 1993; Kirk, Morrow, and Killgore 1994). A final report will be prepared that summarizes our findings and recommends new approaches to monitor triploid grass carp used to control nuisance aquatic vegetation.

Acknowledgments

We thank Dr. John M. Dean of the University of South Carolina for his assistance in locating aging structures. Scott Lamprecht and Miller White of the Freshwater Fisheries Section, South Carolina DNR, greatly assisted us by managing the collection and processing of triploid grass carp.

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Integrated Use of Herbicides and Pathogens for Submersed Plant Control

by

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From an aquatic plant management perspective, integrated control can be defined as a cost-effective, environmentally sound management system that incorporates suitable control techniques (biological, chemical, environmental, physical) to reduce exotic plant populations to levels which cause no economic or ecological harm. This implies a combination of different control measures instead of an approach based on a single control measure (Murphy and Pieterse 1990). The rationale behind this approach is to combine the strengths of each technology which often eliminates their individual weaknesses.

Today the term integrated control is usually associated with techniques aimed at reducing reliance on pesticides. Murphy and Pieterse (1990) stated that in aquatic systems herbicides are only applied if other means of aquatic plant control are ineffective or too costly; therefore, decreasing the use of chemicals is usually not a major objective of integrated control. However, the recent emphasis on using low rates or improving timing of application of herbicides to provide a species-selective response or to reduce environmental loading creates many new potential opportunities to integrate chemicals with other technologies. Herbicide use at reduced rates or the ability to apply less frequently will give plant managers from state and Federal agencies more flexibility in choosing chemicals as part of their plant management program. It is likely that the public will continue to demand a reduction in herbicide usage and use rates. Therefore, it is essential that integrated strategies be developed to meet this demand while maintaining the ability to provide adequate and cost-effective plant control.

Several researchers have investigated combining chemical and biological methods for improved plant control. Efforts directed at submersed vegetation have involved the integration of herbicides with pathogens. Sorsa, Nordheim, and Andrews (1988) examined the effect of combining the herbicide endothall with the fungal pathogen *Colletotrichum* sp. for control of Eurasian watermilfoil. Smit et al. (1990) combined the herbicide fluridone with isolates of various fungal species and evaluated control of coontail. Results suggest the integrated approach provided improved efficacy versus either approach on its own.

Biological control research under the APCRP has identified the fungal pathogen *Mycoleptodiscus terrestris* (MT) as a potential organism to provide control of the exotic submersed species hydrilla (*Hydrilla verticillata* L.f Royle). Moreover, chemical control research under the APCRP has demonstrated that extremely low use rates of the herbicide fluridone can provide effective hydrilla control. Kerfoot (1989) suggested that herbicides that block metabolic pathways (such as fluridone) can weaken the plant defenses at low rates and indirectly increase susceptibility to traditional nonlethal agents such as pathogens.

In assessing the use of either fluridone or MT alone for hydrilla control, it was noted that the strengths of each technology frequently offset the weaknesses of the other technology (Table 1). This assessment led to initial discussions and planning for integrating these technologies in a laboratory-scale demonstration. The objective of this study was to determine the potential additive, antagonistic, or synergistic effects on hydrilla by integrating a fluridone treatment with the plant pathogen MT.

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Table 1
Comparative Strengths and Weaknesses of the Biocontrol Agent MT and the Herbicide Fluridone for Control of Hydrilla

<i>Mycoleptodiscus terrestris</i>	Fluridone
Strengths	Weaknesses
<ul style="list-style-type: none"> • High specificity (studies still required) • Rapid results • Very short exposure requirement 	<ul style="list-style-type: none"> • Low to moderate specificity (species, rate, and timing) • Delayed results • Extended exposure requirement
Weaknesses	Strengths
<ul style="list-style-type: none"> • Variable activity (temperature, virulence) • Little to no field verification • Little residual activity (limitation of current formulation) • Not currently registered for use 	<ul style="list-style-type: none"> • Defined dose response • Proven method of control • No regrowth during exposure • EPA registered

Materials and Methods

This study was conducted in a walk-in environmental growth chamber that has been described in previous herbicide concentration/exposure time studies (Netherland, Green, and Getsinger 1991, Netherland, Getsinger, and Turner 1993). This system consists of 52, 55-L aquaria, located in a controlled-environment room with a temperature of 23 ± 2 °C, light intensity of 570 ± 60 $\mu\text{mol/m}^2/\text{sec}$, and photoperiod of 14L:10D. A general water culture solution proposed by Smart and Barko (1984) for growing aquatic macrophytes was used throughout the study.

Hydrilla apical tips were collected from the Suwannee River, FL, and sediment was obtained from Brown's lake at the WES. Sediment was enriched with NH_4Cl (200 mg/L) to prevent nitrogen limitation during the course of the studies. Glass beakers (300 ml) were filled with sediment and four 10- to 15-cm hydrilla apical tips were planted (5 cm deep) in each beaker. A thin layer of silica sand (0.5 cm) was used to cover the sediment to prevent sediment suspension in the water column. Eleven beakers were placed in each

aquarium and water was flowed-through (one exchange per 24 hr) during the pretreatment growth period. Air was lightly bubbled through each aquarium to provide a source of CO_2 and mixing of the water column.

A pretreatment growth period of 4 weeks resulted in the formation of a thin surface canopy and ensured development of a viable root system. Prior to treatment, one beaker was removed from each aquarium to provide an estimate of pretreatment biomass. Estimated pretreatment shoot biomass (128 g DW m^{-2}) for this study approximates spring-to-early summer biomass reported for hydrilla (Harlan, Davis, and Pesacreta 1985). Treatments were randomly assigned to a test aquarium resulting in 17 exposure scenarios (Table 2). Each treatment was replicated three times.

Two beakers were harvested from each aquarium at 14-, 28-, 42-, 60-, and 94-day posttreatment to determine treatment effects on biomass over time. In addition net photosynthesis (net PHS) and total chlorophyll were measured on 4-cm apical tips throughout the studies to determine physiological competence of treated plants.

Table 2
Fluridone, MT, and Fluridone + MT
Treatment Rates for Control of Hydrilla

Treatment	Rate, $\mu\text{g/L}$	Rate, CFU/ml
Fluridone	2	-
Fluridone	5	-
Fluridone	12	-
MT	-	100
MT	-	200
MT	-	400
Fluridone + MT	2	100
Fluridone + MT	2	200
Fluridone + MT	5	100
Fluridone + MT	5	200
Fluridone + MT	12	100
Fluridone + MT	12	200
Untreated Ref	-	-

Data were analyzed at each sample date using analysis of variance (ANOVA) to determine if differences existed between the various treatments. If the ANOVA determined significant differences existed, data were further subjected to an LSD test at the 0.05 level.

Results and Discussion

Results showed dramatic differences in the pattern of initial and long-term hydrilla response to the various treatments (fluridone, MT, fluridone + MT). As a point of reference, untreated control plants maintained healthy growth throughout the study and physiological parameters (chlorophyll content and net photosynthesis (PHS)) were generally consistent; however, some reduction in vigor was noted (indicating stress or nutrient limitation) toward the end of the study. Following initial treatment injury, if plants began to show physiological recovery, biomass recovery always followed (providing a predictive capability). If, however, physiological parameters remained

significantly reduced compared to untreated controls, hydrilla biomass continued to decline.

Following the fluridone treatments of 2, 5, and 12 $\mu\text{g/L}$, characteristic bleaching of apical tips was noted within days. Overall, hydrilla response to fluridone was dose dependent with significant differences in percent biomass reduction noted at each sample period (Figure 1). This dose response has been noted in previous studies and is especially pronounced at the low fluridone rates tested (Netherland, Getsinger, and Turner 1993). The reduction of biomass was essentially linear over time with eventual reductions of 71, 90, and 97 percent for treatment rates of 2, 5, and 12 $\mu\text{g/L}$, respectively (Figure 1). The inability to completely control hydrilla in the laboratory following extended exposure periods is in agreement with previous fluridone studies (Netherland, Getsinger, and Turner 1993).

Within days following MT treatment, hydrilla leaves became translucent and began to detach from the stems. By 10 days after treatment (DAT), stems were nearly completely defoliated and significant stem damage had occurred at the higher MT treatment rates of 200 and 400 CFU/ml. Initial results indicated that hydrilla also responded to MT treatment in a dose-dependent manner. Chlorophyll and net PHS were significantly reduced compared to untreated controls at 14 DAT. Biomass measurements at 14 DAT indicated reductions of 97, 91, and 82 percent following MT treatments of 400, 200, and 100 CFU/ml (Figure 2). However, physiological parameters measured at 21 DAT began to show recovery in the 100- and 200-CFU/ml treatments. Visual assessments and the 28-day biomass harvest confirmed that although biomass was still significantly reduced compared to untreated controls, recovery had occurred between 14 and 28 DAT. Biomass recovered rapidly and by 42 DAT recovery from 100- and 200-CFU/ml treatments resulted in no significant differences (Figure 2). Although the 400-CFU/ml treatment was generally slower to recover, vigorous growth resulting in canopy formation was noted within 56 DAT. The laboratory response

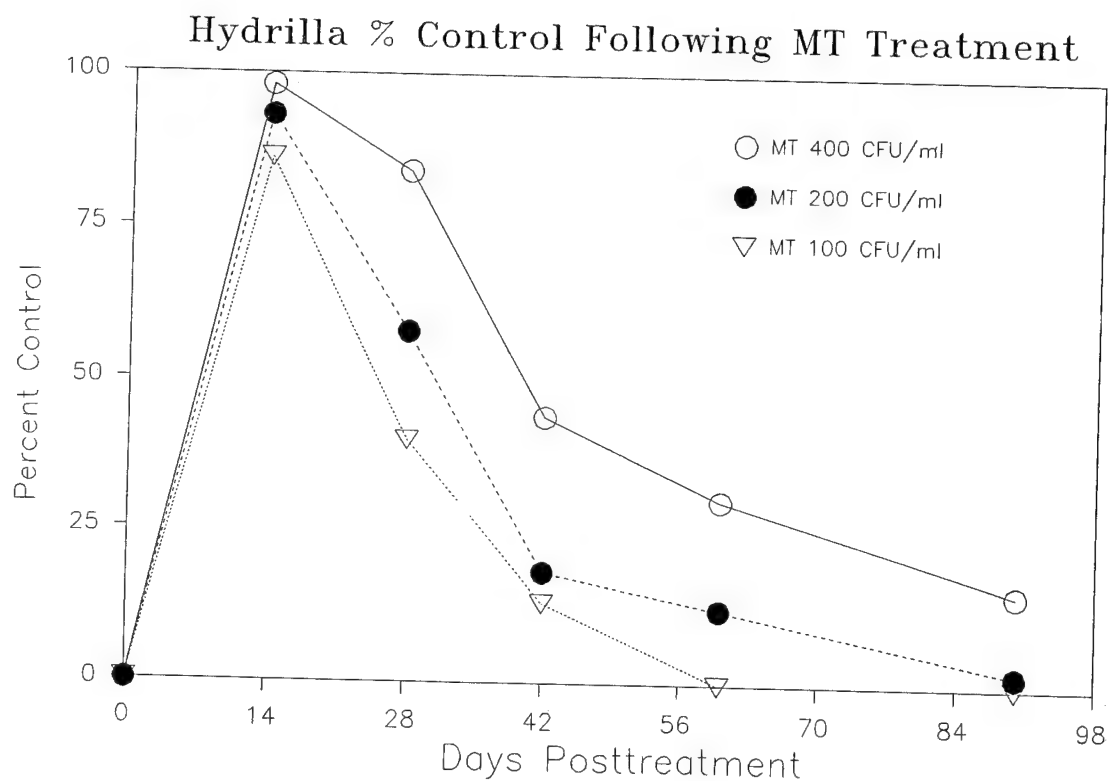
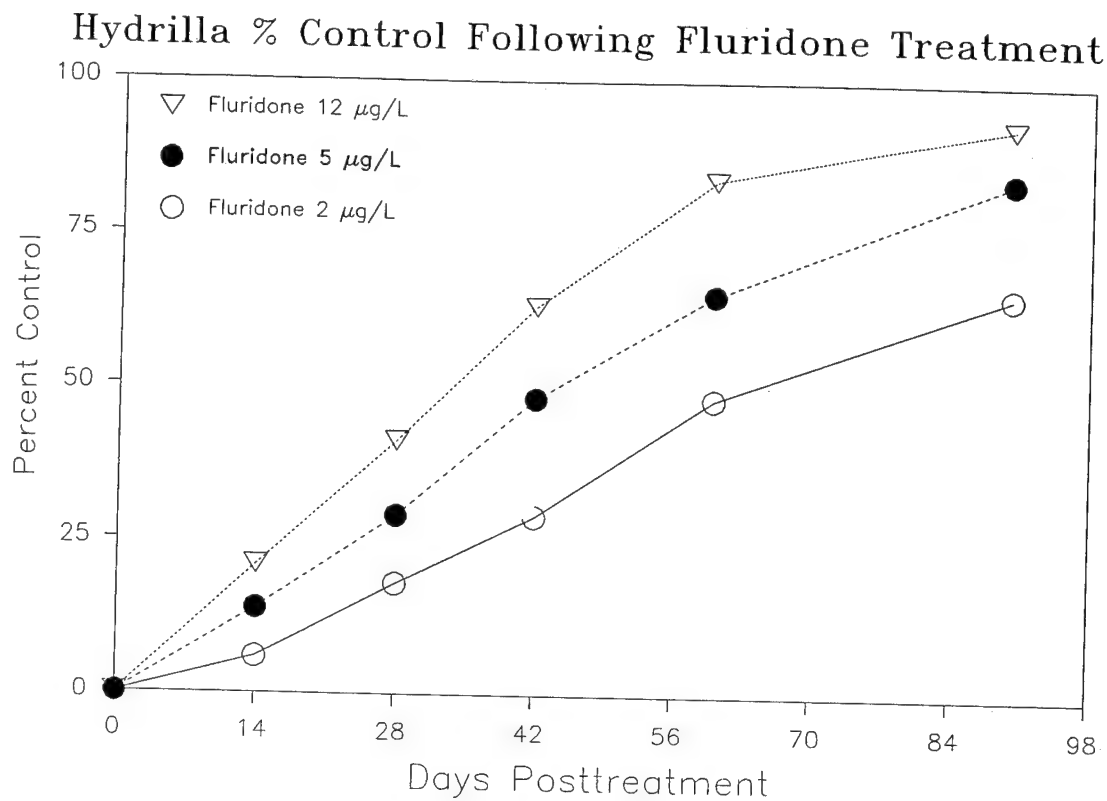


Figure 1. Percent control of hydrilla following treatment with fluridone or *Mycoleptidiscus terrestris* (MT). Points at each sampling date represent the average of three replicate samples

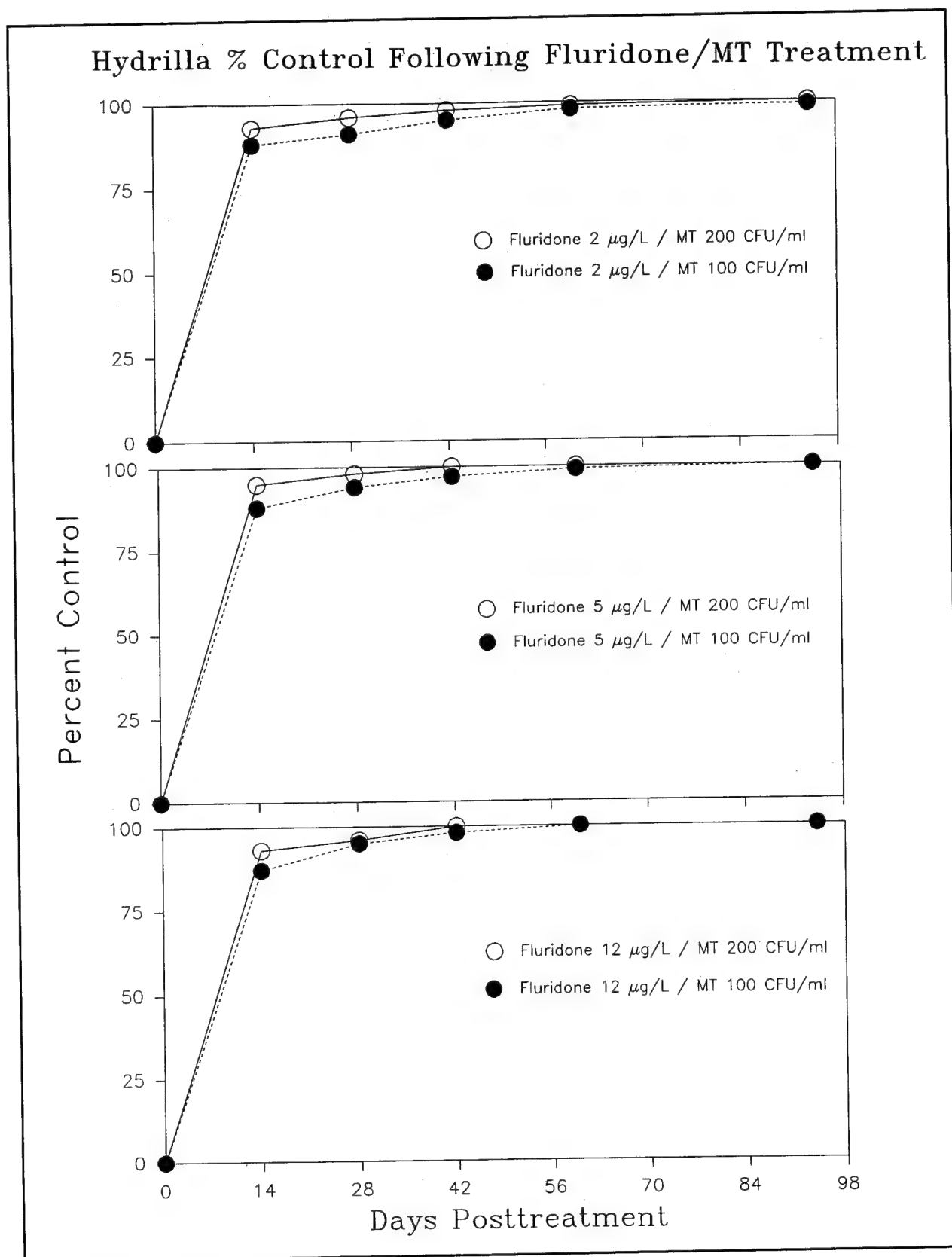


Figure 2. Percent control of hydrilla following treatment with various combinations of fluridone or *Mycoleptidiscus terrestris* (MT). Points at each sampling date represent the average of three replicate samples

of hydrilla to MT treatment (initial knock-down followed by rapid regrowth) was quite similar to effects in pilot-scale field trials at the Lewisville Aquatic Ecosystem Research Facility (LAERF).¹

The fluridone + MT treatments resulted in initial symptoms similar to the MT treatment with leaves becoming translucent and stems defoliated or severely injured (200 CFU/ml) within 1 week posttreatment. By 14 DAT these treatments were indistinguishable from the MT treatments, with biomass reductions ranging from 89 to 96 percent (Figure 2). However, at 21 DAT, physiological parameters showed no signs of recovery. New apical shoots were sprouting from uninjured stems and rootcrowns; however, within days fluridone symptoms (bleached apices) were manifest. Physiological parameters remained significantly reduced compared to untreated controls and biomass continued to decline. This eventually resulted in complete hydrilla control at 56 and 42 DAT with the 5 and 12 µg/L + 100- and 200-CFU/ml treatments. The 2-µg/L fluridone + MT at 200 and 100 CFU/ml resulted in complete control of hydrilla by 60 to 94 DAT.

Laboratory results suggest that MT (as currently formulated) acts as a contact bioherbicide which results in excellent initial biomass reduction, but fails to provide any long-term residual control. (It should be noted that the WES biocontrol team is currently working on formulating MT to provide improved residual control.) Fluridone treatment provided good long-term hydrilla control; however, poor initial control and the requirement for extended exposure periods limit applications of this product. (It should also be noted that the WES chemical control team is evaluating delivery systems (controlled-release, metering, repeat applications) to optimize the use of fluridone.)

Integrating fluridone and MT provided the benefits of excellent initial biomass reduction along with long-term control. Integrating these treatments greatly reduced fluridone exposure requirements while also reducing the

rate of fluridone necessary to provide control of hydrilla.

Preliminary laboratory results demonstrate the potential exists for combining chemical and biological control agents to improve efficacy and reduce reliance on chemicals (lower use rates or less frequent applications). In addition, by assessing the comparative strengths and weaknesses of a chemical with a biological control agent, a rational approach can be used to combine these technologies.

Future Work

Future work will include determination of the specificity (hydrilla and beneficial native species) of both MT and fluridone individually and in combination. Studies will be scaled up to the mesocosm level at the LAERF for validation of laboratory results and to determine the effects of such variables as temperature, water quality, and species composition on various fluridone and MT treatment combinations. In addition, following operational field treatments, hydrilla will be analyzed in the laboratory to determine pathogenic assemblages associated with a fluridone treatment.

Acknowledgments

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The Influence of an Exotic Submersed Aquatic Plant, *Myriophyllum spicatum*, on Water Quality, Vegetation, and Fish Populations of Kirk Pond, Oregon

by

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Kirk Pond, located near Eugene, OR, is a 24-ha borrow pit resulting from the construction of the Fern Ridge Reservoir dam. It is the central feature of a day-use park, with primary activities being fishing and nonpower boating. The primary water source is a pipe from Fern Ridge Reservoir. The outlet of Kirk Pond, Coyote Creek, is a tributary of the Long Tom River. The nonnative submersed macrophyte *Myriophyllum spicatum* (Eurasian watermilfoil) currently dominates the pond, interfering with recreation and thus reducing the abundance and diversity of both native plants and fish. This article documents environmental and ecological information required to evaluate the actual and potential effects of Eurasian watermilfoil on the pond ecosystem and the need for aquatic plant control.

chemistry analytical procedures are described in Madsen et al. (1993).

The pond can be delineated into two major areas by water quality and flow regime. The western basin of the pond is essentially a flow-through system for outlet water from Fern Ridge Reservoir. The eastern basin of the pond is an isolated water body, relatively static, and appears to have significant influx of groundwater or seepage water. In general, the eastern basin has higher conductivity, alkalinity, and dissolved metals, and lower turbidity and suspended solids than the western basin (Table 1). Turbidity is a major factor in vegetation distribution and abundance in the pond, as indicated later (Figure 1).

Characterization

Bathymetry and sediment composition were measured along transects established every 400 m along the shoreline of Kirk Pond. Sediment analyses were described in Madsen et al. (1993). Water chemistry was measured at six stations in the pond every two weeks during May through October. Stations 1 and 6 were grab samples from the inlet and outlet, respectively (Figure 1). Stations 2 through 5 were collected as integrated samples of the water column. Water

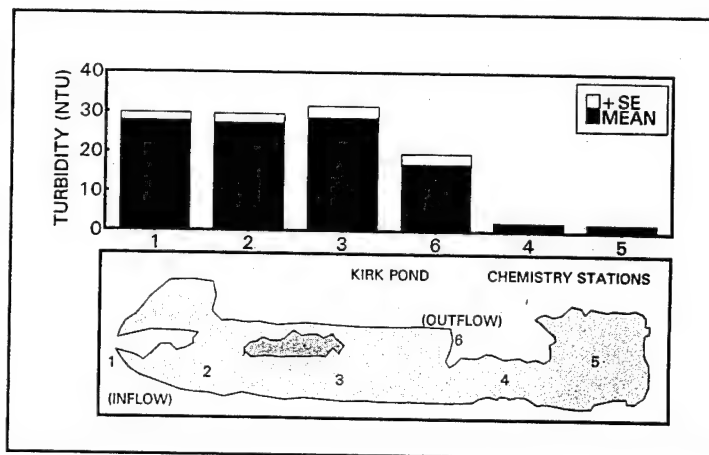


Figure 1. Seasonal average turbidity of water samples from all sample dates at the six water chemistry sample sites (top), and location of water sample stations in Kirk pond (bottom)

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Table 1
Water Chemistry Parameters for Six Monitoring Stations in Kirk Pond Averaged
Over the Entire Sampling Period. (Standard error of the mean is given in
parentheses)

Station: Parameter	1 Inflow	2 In-lake	3 In-lake	4 In-lake	5 In-lake	6 Outflow
Temp (°C)	19.4 (0.3)	19.4 (0.7)	20.0 (0.9)	20.5 (0.9)	20.1 (0.8)	20.0 (0.7)
pH (units)	7.1 (0.2)	7.3 (0.1)	7.7 (0.1)	9.4 (0.2)	9.6 (0.3)	8.1 (0.2)
D.O. (mg L ⁻¹)	7.6 (0.4)	8.2 (0.4)	8.6 (0.4)	10.4 (0.8)	8.4 (0.5)	7.9 (0.6)
Cond. (μS cm ⁻¹)	79.7 (8.2)	75.0 (9.2)	83.3 (8.4)	184.8 (21.3)	194.0 (18.1)	81.2 (10.0)
Alk. (mg CaCO ₃ L ⁻¹)	24.0 (1.4)	25.9 (1.5)	29.7 (1.4)	91.1 (2.5)	93.6 (2.8)	29.2 (1.3)
Turb. (NTU)	27.8 (1.9)	27.3 (2.2)	28.6 (2.9)	2.1 (0.2)	1.9 (0.2)	17.1 (2.6)
TSS (mg L ⁻¹)	22.8 (2.8)	21.6 (1.9)	38.0 (13.0)	7.7 (1.8)	4.7 (1.6)	18.7 (3.2)
NH ₄ -N (mg L ⁻¹)	0.03 (0.01)	0.08 (0.04)	0.06 (0.03)	0.04 (0.01)	0.03 (0.02)	0.03 (0.01)
NO ₃ -N (mg L ⁻¹)	0.05 (0.01)	0.03 (<0.01)	0.02 (<0.01)	0.03 (0.01)	0.03 (0.01)	0.03 (0.01)
TP (μg L ⁻¹)	48 (4)	58 (6)	60 (9)	26 (2)	20 (2)	40 (3)
SRP (μg L ⁻¹)	6 (0.3)	8 (0.6)	8 (0.1)	10 (0.9)	4 (0.6)	7 (0.9)
Ca (mg L ⁻¹)	2.8 (0.2)	3.3 (0.3)	3.9 (0.3)	13.9 (0.7)	13.8 (0.5)	4.3 (0.5)
Mg (mg L ⁻¹)	2.5 (0.1)	2.9 (0.1)	3.5 (0.3)	12.6 (0.5)	12.8 (0.3)	3.4 (0.2)
Na (mg L ⁻¹)	4.0 (0.4)	4.3 (0.3)	4.7 (0.3)	9.7 (0.8)	10.5 (0.9)	4.7 (0.2)
K (mg L ⁻¹)	1.1 (0.1)	1.3 (0.2)	1.0 (0.1)	0.9 (0.1)	1.0 (0.1)	0.9 (0.0)

Vegetation—Distribution

The distribution of aquatic plants in Kirk Pond was examined using transects placed every 400 m, marked in 1-m intervals. All species were recorded in each 1-m interval by a SCUBA diver. A total of 19 transects and 3,161 intervals were examined. Fourteen

aquatic plant species were observed (Table 2, Figure 2). Total plant cover of the pond was 85 percent. Eurasian watermilfoil covered a total of 69 percent of the pond, with 50 percent being a monoculture of *M. spicatum*. The 15 percent bare area occurred in two deep spots (exceeding 1.5 m) in the turbid western basin of the pond. Native plants,

Table 2
Percent Cover of Aquatic Plant Species in Kirk Pond

Scientific Name ¹	Common Name	Growth Form	Percent Cover
<i>Ceratophyllum demersum</i>	Coontail	Submersed	24.7
<i>Cyperus</i> sp.	Rush	Emergent	0.4
<i>Elodea canadensis</i>	Elodea	Submersed	9.0
<i>Juncus</i> sp.	Rush	Emergent	1.0
<i>Myriophyllum spicatum</i>	Eurasian watermilfoil	Submersed	68.9
<i>Nuphar</i> sp.	Yellow pond lily	Floating-leaved	0.1
<i>Polygonum</i> sp.	Smartweed	Emergent	1.1
<i>Potamogeton nodosus</i>	American pondweed	Floating-leaved	0.3
<i>Potamogeton crispus</i>	Curlyleaf pondweed	Submersed	0.5
<i>Potamogeton epihydrus</i>	Leafy pondweed	Submersed/Floating-leaved	0.1
<i>Scirpus</i> sp.	Bulrush	Emergent	1.5
<i>Sparganium</i> sp.	Burreed	Emergent	0.9
<i>Typha</i> sp.	Cattail	Emergent	1.0
<i>Utricularia vulgaris</i>	Common bladderwort	Emergent	0.3
None (Bare Sediment)	--	--	15.0

¹ Species identified to generic level only because of the absence of flowering structures or other identifying features required for proper identification.

without any Eurasian watermilfoil, constituted only 16 percent of the pond and were largely

restricted to a deep area in the eastern basin of the pond (Figure 2). This deep area was dominated by *C. demersum* (coontail), a native species that tolerates low light levels. Diversity throughout the pond was relatively low, in part due to the dominance of much of the habitable space by *M. spicatum*. Native species occurred in 51 percent of the intervals not inhabited by *M. spicatum*, and only 27 percent of intervals which did have Eurasian watermilfoil (Table 3).

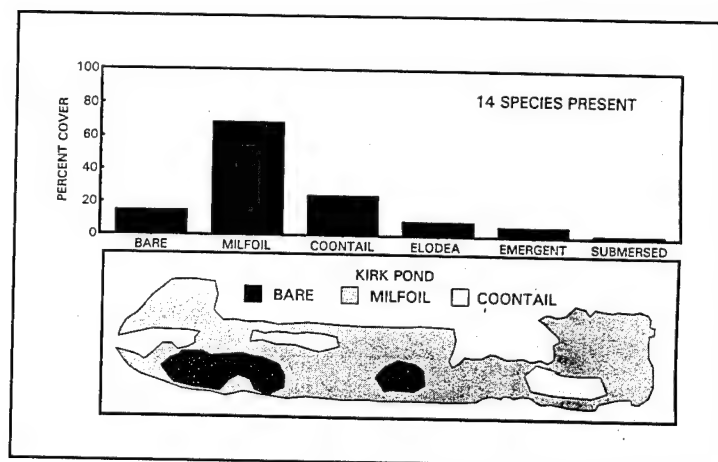


Figure 2. Percent surface area of Kirk Pond composed of bare substrate, milfoil, coontail, elodea, emergent species, and other submersed species (top) and distribution map of major plant communities in Kirk Pond (bottom)

Vegetation—Biomass

In addition to limiting the depth distribution of plants in the western end of the pond, turbidity also appeared to limit the biomass of *M. spicatum*. Biomass at stations in the eastern basin of the pond was greater than 300 g m⁻²

Table 3
Percent Frequency of Plant Species Pairwise Two-by-Two Comparisons of Presence or Absence in the Presence or Absence of *Myriophyllum spicatum*. (Species with too few occurrence for statistical comparisons were eliminated)

Species	Without <i>M. spicatum</i>		With <i>M. spicatum</i>		Chi-sq p-value and +/- Association
	Absent	Present	Absent	Present	
<i>Ceratophyllum demersum</i>	543 (55)	441 (45)	1838 (84)	339 (16)	0.001 -
<i>Elodea canadensis</i>	918 (93)	66 (7)	1959 (90)	218 (10)	0.003 +
Native plants ¹	478 (49)	506 (51)	1582 (73)	595 (27)	<0.001 -

¹Cross-classification based on the occurrence of any native species in the presence or absence of *M. spicatum*; thus it excludes both *M. spicatum* and *P. crispus*.

and appeared limited by nutrient availability. Biomass levels in the western basin were below 300 g m⁻², and were not limited by nutrient availability. Biomass of *M. spicatum* in the western basin of the pond may have been limited by light due to the higher turbidity in this area (Figure 3; Madsen et al. 1993).

Fisheries

Kirk Pond was sampled in the spring of 1991 and 1992 using electroshocking. A total of eleven species were collected, with bluegill and white crappie being the most common species (Table 4). The size distribution of

the three major fish species indicated poor growth in the dense cover created by Eurasian watermilfoil (Figure 4).

Conclusions

Kirk Pond was dominated by a dense canopy of *Myriophyllum spicatum* that reduced native plant diversity, reduced the growth and vigor of the warm-water fishery, and interfered with the recreational use of the pond. With a reduction in turbidity, Eurasian watermilfoil could spread to deeper areas of the western basin of the pond, as in the eastern basin.

Areas of more than 1.5-m depth in the western basin that are currently bare might be colonizable by coontail. Without management of *M. spicatum*, the abundance and diversity of native plants in the 0.5- to 2.0-m depth range will likely continue to decline. Management of *M. spicatum* will improve native plant diversity and abundance, growth of warm-water fishes, and access for recreational purposes.

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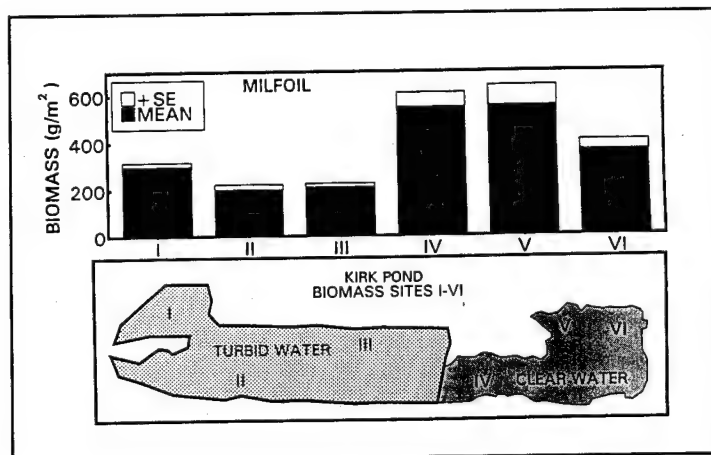


Figure 3. Biomass of *Myriophyllum spicatum* at six biomass sample locations (top) and locations of biomass sample sites and demarcation of turbid water and clear water regions of Kirk Pond (bottom)

Table 4

Species List and Frequencies from 1991 and 1992 Kirk Pond Electrofishing Samples. (*f* is the number of each species collected and *rf* is the relative frequency within all samples)

Family	Scientific Name	Common Name	1991		1992	
			<i>f</i>	<i>rf</i> (%)	<i>f</i>	<i>rf</i> (%)
Salmonidae	<i>Salmo clarki</i>	Cutthroat trout	6	1.8	--	--
Cyprinidae	<i>Carassius auratus</i>	Wild goldfish	--	--	1	0.3
	<i>Cyprinus carpio</i>	Common carp	13	4.0	29	9.4
Catostomidae	<i>Catostomus macrocheilus</i>	Largescale sucker	1	0.3	1	0.3
Ictaluridae	<i>Ictalurus natalis</i>	Yellow bullhead	2	0.6	--	--
Poeciliidae	<i>Gambusia affinis</i>	Mosquitofish	*	*	*	*
Centrarchidae	<i>Micropterus salmoides</i>	Largemouth bass	21	6.4	17	5.5
	<i>Lepomis gibbosus</i>	Pumpkinseed	4	1.2	1	0.3
	<i>Lepomis gulosus</i>	Warmouth	17	5.2	5	1.6
	<i>Lepomis macrochirus</i>	Bluegill	126	38.7	51	16.5
	<i>Poxomis annularis</i>	White crappie	126	38.7	202	65.2
	<i>Poxomis nigromaculatus</i>	Black crappie	10	3.1	3	0.9
		Totals	326	100.0	310	100.0

* Collected only during preliminary sampling.

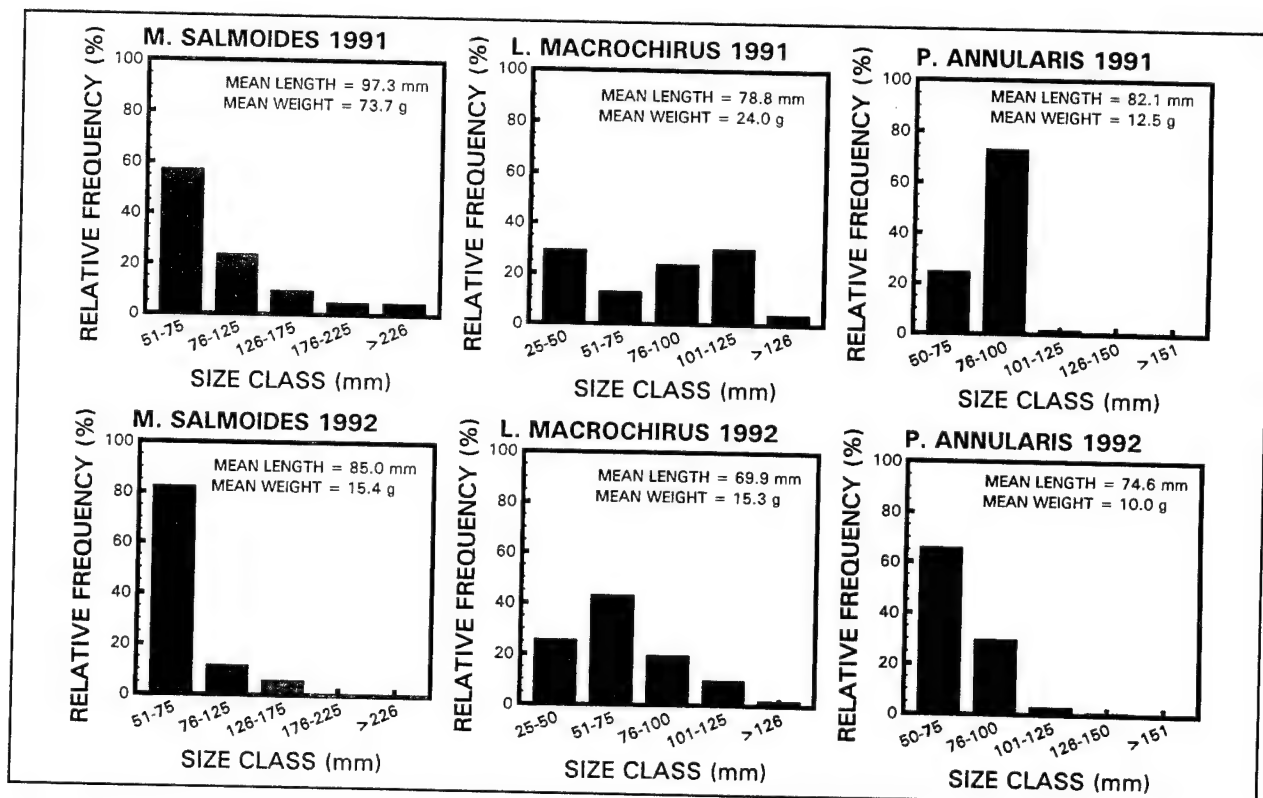


Figure 4. Size class distributions of largemouth bass (*M. salmoides*), bluegill (*L. macrochirus*), and white crappie (*P. annularis*) for 1991 and 1992, with mean length and weight also indicated

Synopsis of the District/Division Aquatic Plant Management Operations Breakout Session

by
Wayne T. Jipsen¹

The eighth annual Operations Breakout Session was held on 15 November 1994 during the Aquatic Plant Control Research Program Review. Representatives from Headquarters, 1 Division Office, 10 District Offices, and the WES attended. The total attendance of 27 individuals also included representatives from 7 state cooperating agencies.

The session started with an overview of the program provided by the Headquarters' APC Program Manager. The major topics of discussion during this overview were funding concerns and the revision of the Corps APC Regulation. The revision is part of the Federal government's overall program to reduce regulations. The format of the new regulations allows for the description of the program but does not include the customary "processes." These will be dictated later as policy.

Each District and local sponsor representative briefed the meeting attendees on their re-

spective programs. Three main topics emerged from these overviews. First, all major submersed exotic species currently targeted in the program are expanding their ranges. Hydrilla continues to spread in Alabama, Florida, North Carolina, South Carolina, and Texas. Eurasian watermilfoil is spreading in Michigan, Minnesota, Tennessee, and Wisconsin while water chestnut is spreading in the Northeast.

As a result of this spread, the second main concern dealt with the desire to expand existing programs and/or initiate new starts. Headquarters guidance was to document the needs and send the requests forward for review.

Lastly, more states are utilizing dedicated funding sources (taxes and/or fees) for their local sponsor share. This approach is getting mixed results around the country.

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Chemical Control Technology

Chemical Control Technology Development: Overview

by

Kurt D. Getsinger¹

The mission of the Chemical Control Technology Development area is to develop and evaluate technology that will improve the management of nuisance aquatic vegetation using herbicides and plant growth regulators (PGRs) in an environmentally compatible and cost effective manner. A wide array of work is being conducted in laboratory and growth chamber facilities at WES, in large-scale outdoor mesocosms at LAERF near Dallas, TX, and at various field sites around the Nation. In fiscal year 1994 (FY94), direct allotted funds for chemical control research and development were apportioned among six work units:

- a. Herbicide Concentration/Exposure Time Relationships.
- b. Herbicide Application Techniques for Flowing Water.
- c. Herbicide Delivery Systems.
- d. Field Evaluation of Selected Herbicides.
- e. PGRs for Aquatic Plant Management.
- f. Species-Selective Use of Aquatic Herbicides and PGRs.

Two additional work units, (g) Coordination of Control Tactics with Phenology of Aquatic Plants and (h) Integrated Use of Herbicides and Pathogens for Submersed Plant Control, were also under the direction of the Chemical Control Technology Team (CCTT) during FY94.

For the past several years, the CCTT's program has continued to focus on the protection and restoration of aquatic habitats using herbicides and PGRs. The emphasis of this restoration effort involves the selective removal of

nuisance plants, allowing for the growth and recovery of native vegetation (Figure 1). Through the use of selective chemical control techniques, biodiversity of a system can increase while dependence on herbicides (and other control tactics) can decrease. This management scenario will allow for less environmental loading of chemicals, as well as reduced labor and costs associated with repetitive herbicide application techniques.

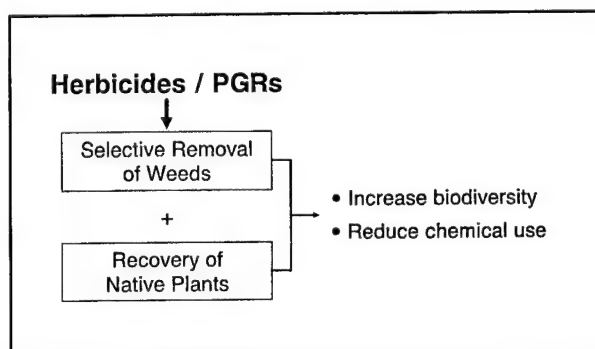


Figure 1. Protection/restoration of aquatic habitats using chemicals

Cooperative work in the chemical area includes efforts with various CE Districts (e.g., Jacksonville, Seattle, St. Paul, and Wilmington), TVA, USDA, USDI-BR, state and local entities (e.g., Washington Department of Ecology, Minnesota Department of Natural Resources, Texas Parks and Wildlife, and Florida Water Management Districts), and research institutions (e.g., University of Florida, Purdue University, and North Carolina State University). The CCTT also interacted with chemical companies pursuing aquatic registration for new products earmarked for use in public waters and/or changes in existing national aquatic herbicide labels (e.g., DowElanco,

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

SePRO, Griffin, Rhone-Poulenc, Elf Atochem, Zeneca).

In an effort to communicate more effectively with the pesticide regulatory arm of the USEPA, the CCTT established an AHWG comprised of representatives from key Federal agencies involved in studying, regulating, and managing aquatic vegetation: the CE, USDA, TVA, USDI-BR, and USEPA. This working group will operate under the sponsorship of the CAPRT, which was recently established at the WES. Functions of the AHWG are as follows:

- a. Serve as clearinghouse for all Federal agencies implementing chemical control of aquatic vegetation.
- b. Coordinate Federal research efforts to ensure the development of safe and cost-effective herbicides, and provide guidance for the environmentally sound use of those compounds in public waters.
- c. Address issues concerning the fate, persistence, and toxicity of herbicides in the aquatic environment.
- d. Address issues concerning the potential adverse human health effects of aquatic herbicides, including applicator safety.
- e. Interact, when appropriate, with state and local agencies tasked with managing aquatic vegetation.
- f. Encourage research initiatives in government, academia, and private industry that focus on the integration of chemical control methods with other aquatic plant management technologies.
- g. Provide public information on the risks and benefits associated with the appropriate use of aquatic herbicides.

The AHWG will be chaired by the CCTT, and will meet on a regular basis to discuss items of mutual interest concerning chemical control of aquatic vegetation.

Chemical control work unit summaries are presented below. Detailed updates of each work unit can be found in articles in the Chemical Control Technology Section of this proceedings.

Herbicide Concentration/Exposure Time Relationships (32352)

Concentration/exposure time (CET) relationships are being developed for all registered and experimental use permit herbicides used for controlling Eurasian watermilfoil and hydrilla. Unique properties of each chemical (i.e. application rate, mode of action, half-life, and selectivity) require that CET relations be developed for each target plant. These CET studies are conducted in controlled-environment chambers at WES and in outdoor mesocosms at the LAERF. When coupled with water exchange information, CET relationships can be used to guide the selection, rate, and timing of application of appropriate herbicides. When herbicides are used within a respective CET window for target plant control, concomitant effects on non-target submersed plants are also being ascertained in small-scale evaluations.

Herbicide Application Techniques for Flowing Water (32354)

In this effort, herbicide application techniques are developed to minimize the amount of active ingredient used and the frequency of treatments, while maximizing efficacy against submersed target plant species (primarily Eurasian watermilfoil and hydrilla) in high water exchange environments (e.g., rivers, canals, tidal zones, lakes, and reservoirs). Results from the previously described Herbicide CET work unit provide the pertinent dose/response information required to develop improved and innovative application techniques.

Work has focused on understanding bulk water exchange and stratification patterns in target submersed plant stands, and on the potential impact of these conditions on herbicide contact time, off-target movement, and efficacy. Water exchange studies have been completed using tracer dyes and flow meters in a variety of submersed plant stand conditions. Results from this work are being used by operational personnel to select both type and timing of submersed application techniques that will minimize the impacts of water movement on

herbicide efficacy. Successful large-scale applications of products developed in this work unit include the control of hydrilla in the Crystal, Withlacoochee, and St. Johns rivers, FL, and in Foster Creek, SC, using the herbicides endothall and fluridone; and the control of Eurasian watermilfoil in Pend Oreille River and Long Lake, WA, and in Lake Minnetonka, MN, using the herbicides triclopyr and fluridone. This flowing water work unit is scheduled for completion in FY95.

Herbicide Delivery Systems (32437)

Information generated in the Herbicide CET and Flowing Water work units, above, is used to design systems that can deliver low doses of herbicides over extended periods of time, providing acceptable plant control. Environmentally compatible controlled-release (CR) carriers, such as polymers, gypsum, proteins, etc., are evaluated for herbicide release rates and efficacy in small-scale systems at WES. The most promising formulations are further evaluated in large-scale mesocosms at the LAERF, or in hydraulic channels at the TVA Aquatic Research Laboratory near Athens, AL. Results from these studies are used to improve the control of nuisance submersed plants in flowing water systems. Recent work has focused on endothall-gypsum CR carriers and a high-load endothall-polymer formulation.

Field Evaluation of Selected Herbicides for Aquatic Use (32404)

The most effective application techniques and chemical formulations developed through the CCTT's small-scale research are eventually evaluated under field conditions. In addition to documenting efficacy, these field efforts are used to collect pertinent environmental data pertaining to the fate and dissipation of the chemical formulations' active ingredients. Cooperators on these efforts include chemical companies, CE Districts, Federal, state, and local agencies, universities, and contractors. Efficacy and environmental fate and dissipation data collected in these studies are used to

prepare guidance on the appropriate use of aquatic herbicides, and are available to support USEPA requirements for registration of specific herbicide formulations for use in public waters. Results from these field evaluations verify results from laboratory- and mesocosm-scale studies, and can aid in modifying the status of previously registered aquatic herbicides. Recent work has focused on the national aquatic registration of triclopyr (for Eurasian watermilfoil and waterhyacinth control), and on modifying the labels on fluridone and endothall.

Plant Growth Regulators for Aquatic Plant Management (32578)

Plant growth regulators (PGRs) offer the potential of slowing the vertical growth rate of nuisance submersed plants, thereby reducing the negative aspects that "topped-out" vegetation can impose on a water body (e.g., restricting navigation and recreation, causing wide diurnal variance in water temperature, dissolved oxygen, and pH levels, and eliminating native plant habitat). Thus, the beneficial qualities provided by underwater vegetation (e.g., fish and wildlife habitat, oxygen production, nutrient sink, and sediment stabilization) can be retained.

Chemical compounds that demonstrate PGR activity are being evaluated in this work unit including bensulfuron methyl, flurprimidol, paclobutrazol, and uniconazole. Since PGR efficacy is sensitive to life cycle events of target plants, information obtained from the Phenology work unit (described below) will be useful in evaluating growth regulating effects. Once evaluations have been conducted in greenhouse and growth chamber systems, the most promising compounds are evaluated in mesocosms and ponds at the LAERF.

Species-Selective Use of Aquatic Plants and PGRS (32841)

While weedy submersed species can be removed using traditional chemical control tactics, these treatments can also impact native

plant species. However, using chemicals in a species-selective manner can result in the control of target vegetation while enhancing the growth of desirable/beneficial plants. Allowing these desirable species to flourish can slow the reinvasion of weedy species and provide improved fish and wildlife habitat. In this way, water bodies plagued with monoculture infestations of exotic plants can be restored to a healthy, diversified, and balanced aquatic community.

In this work unit, species-selective aquatic plant management practices using chemicals are being developed and evaluated. Studies are focusing on species-selective responses to applications (rates and timing) of various herbicides/PGRs. As responses to weedy and various nonweedy species are determined, desirable herbicide-resistant plants can be selected for further evaluation. The most promising compounds (e.g. triclopyr, fluridone, endothall, diquat) are being applied to mixed plant communities in a mesocosm system at the LAERF. Results from this effort will be used to provide guidance for managing aquatic vegetation using the species-selective approach.

Coordination of Control Tactics with Phenological Events of Aquatic Plants (32441)

A thorough understanding of a species' survival strategy can be used to identify weak points in its growth cycle, which can then be exploited to improve control of that plant. Once identified, these susceptible periods can be predicted on the basis of growth-cycle events, morphological characteristics, and environmental cues. An easily recognizable characteristic or cue will enable field personnel to determine the optimum time for applying appropriate chemical control techniques

by taking advantage of the weak link in the plant's growth cycle to maximize efficacy.

Phenological studies are being conducted on Eurasian watermilfoil, hydrilla, and waterhyacinth at the LAERF. Results from these studies are being used in the control oriented chemical work units described above. In addition, phenological information is contributing to the plant growth modeling effort in the Simulation Technology area of the APCRP.

Integrated Use of Herbicides and Pathogens for Submersed Plant Control (32953)

Current policies are directing government agencies to reduce overall use of pesticides, including herbicides used for managing nuisance vegetation. One potential method of reducing herbicide use rates while maintaining or improving plant control is to integrate chemical control strategies with known pathogens. By integrating the strengths of a chemical treatment with a biological organism, the weaknesses of an individual control technique are often minimized or negated. Potential uses of herbicide/pathogen combinations include: (1) using low rates of herbicide to stress target plants, making them more susceptible to pathogenic attack, and (2) use of pathogens as contact bioherbicides to reduce standing biomass of target plants which allows for reduced rates of herbicides to be applied.

Selected aquatic herbicide and pathogen combinations are being evaluated for synergistic/antagonistic relationships in growth chamber studies. Promising combinations will be evaluated in mesocosms and ponds at the LAERF. Results from this work will provide information on the potential of integrating chemical and biological control techniques to improve management of aquatic vegetation.

Herbicide Application Technique Development for Flowing Water: Summary of Accomplishments (32354)

by
Kurt D. Getsinger¹

Introduction

Since the major pathway for herbicide uptake in submersed plants is via the shoot/water column interface, hydrodynamic processes can dramatically influence the efficacy of a treatment by altering herbicide concentration and exposure time relationships with respect to target vegetation. Undoubtedly, many unsuccessful chemical treatments can be attributed to off-target movement of water caused by flow, wind mixing, thermal stratification, tidal action, and other water exchange processes. Clearly, an understanding of water movement in and around target submersed vegetation was essential to improve control of nuisance plants in flowing water situations, or in sites representing spot-treatment applications and partial treatments of large water bodies.

In 1986, this "flowing water" work unit was initiated to (a) characterize flow velocities and water exchange patterns in submersed plant stands under a variety of field conditions, (b) evaluate application techniques that maximize herbicide contact time in flowing-water environments, and (c) provide guidance to operational personnel for improving the control of nuisance submersed vegetation using herbicides in high water exchange environments. These work unit objectives were met through a series of large-scale water exchange studies (flow meter and tracer dye) and field application technique evaluations (dye/herbicides). Results from this 10-year effort have been used to dramatically improve the chemical control of Eurasian watermilfoil and hydrilla in rivers, lakes, and reservoirs around the Nation. In addition, treatment techniques developed in this work unit can be used to manage other target submersed species.

The following article presents a summary of the major accomplishments in this work unit including the conduct of water exchange studies to develop and evaluate innovative application techniques, and the verification of those techniques using several herbicides, against two target species, in a variety of field situations. A final summary report on this effort will be available in 1996.

Results and Discussion

Flow meter studies: 1986-87

Flow velocities were measured in submersed plant stands using electromagnetic sensor portable velocity meters in hydraulic flumes at the WES, in the Holston River, TN, and in irrigation/drainage canals in the Sacramento Valley, CA (Getsinger 1987, 1988; Getsinger, Green, and Westerdahl 1990). Results from this work verified that dense stands of submersed macrophytes substantially alter water movement in lotic systems. The physical structure and height of plants, as well as areal extent of the plant stand, can influence water velocities. Data showed that electromagnetic flow meters can be used to characterize linear water flow in and around submersed plant stands when velocities are >1 cm/sec; however, low intrastand velocities (<1 cm/sec) can make the use of these instruments impractical in many field situations. The limited value of these flow meters for determining water exchange patterns in plant stands led to the search for more reliable techniques. One such technique is the use of the inert tracer dye, rhodamine WT (RWT).

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

Dye studies: 1987-90

In an effort to more accurately characterize flow velocities and water exchange patterns in submersed plant stands, field studies were designed to include the use of the dye rhodamine WT. Data from these dye studies were also used to evaluate various submersed application techniques.

The research focused on several major problems faced by field personnel responsible for controlling submersed vegetation. Studies were designed to determine retention time of herbicides in hydrilla stands in tidally influenced systems, such as the Crystal River, FL, and the Potomac River near Washington, DC (Fox, Haller, and Getsinger 1991, Fox et al. 1991; Getsinger et al. 1991). Additional studies were conducted in hydrilla and Eurasian watermilfoil stands in inland rivers and lakes, such as the St. Johns and Withlacoochee rivers, Lochloosa Lake, and Lake Kissimmee in Florida (Getsinger, Green, and Westerdahl 1990; Getsinger, Haller, and Fox 1990), and the Pend Oreille and Columbia rivers in Washington (Getsinger, Green, and Westerdahl 1990; Getsinger, Sisneros, and Turner 1993).

Results of these studies showed that RWT dye is a reliable and relatively easy technique for determining water exchange characteristics, relative to potential herbicide contact time, in submersed plant stands under most field conditions. Results also showed that tracer dyes can be used to determine the effects of thermal stratification and various application techniques on the potential distribution of herbicides in submersed plant stands.

Dye/herbicide studies and operational successes: 1989-95

The primary objective of the final phase of the flowing water work unit was designed to couple field water exchange information, via dye applications, with laboratory-derived herbicide concentration/exposure time (CET) relationships to improve control of target plants in flowing water systems. A secondary objective of this effort was to establish corre-

lations between the behavior of RWT and selected herbicides when applied to submersed plant stands. Once established, these dye/herbicide relationships could be used to reliably predict posttreatment water-column distribution, off-target movement, and contact time of herbicide active ingredients.

Field studies in which RWT was added concurrently to herbicide applications on hydrilla were conducted for diquat, endothall, and fluridone, and bensulfuron methyl in Lakes Orange, Washington, Hell 'n' Blazes, and Seminole, and the Crystal River in Florida (Fox, Haller, and Shilling 1991; Fox, Haller, and Getsinger 1992, 1993; Fox and Haller 1994; Langeland et al., in press). Additional dye/herbicide studies were conducted in Eurasian watermilfoil stands in the Pend Oreille River, WA (Getsinger, Turner, and Madsen 1992a, 1992b, 1993, 1994; Turner, Getsinger, and Netherland 1994).

Results from these evaluations verified herbicide CET relationships for hydrilla and Eurasian watermilfoil, established correlations between the behavior of RWT and most of the registered aquatic herbicides, and contributed to the development of innovative application techniques (e.g., sequential/block treatments, controlled-release carriers, low-dose exposures, etc.).

An important legacy of the flowing water work unit has been the documented operational success stories of managing vegetation in rivers, canals, lakes, and reservoirs that have occurred through the use of the technology developed in this effort (Haller, Fox, and Shilling 1990; Fox and Haller 1992; Getsinger, Turner, and Madsen 1992a, 1993; Farone and McNabb 1993; Getsinger 1993; Fox, Haller, and Shilling 1994; Madsen, Getsinger, and Turner 1994; Getsinger 1995; Madsen and Getsinger 1995). Clearly, the ability to control nuisance submersed plants using herbicides in hydrodynamic environments has been vastly improved by coupling water exchange information, herbicide CET relationships, and innovative application techniques.

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Plant Growth Regulators for Aquatic Plant Management

by

Carole A. Lembi,¹ Sally L. Yost,¹ and Xiao-xi Liu¹

Introduction

For several years we have studied the potential of several groups of chemicals to act as plant growth regulators on submersed aquatic plant species. One of the most promising of these groups is the gibberellin synthesis (GS) inhibitors. These compounds inhibit the synthesis of the naturally occurring plant hormone gibberellin. Since gibberellin is responsible for causing plants to increase in length, application of GS inhibitors reduces main stem elongation. The aquatic plants remain short and nonweedy, and yet are viable and able to provide benefits to the aquatic environment such as oxygen production and habitat.

We now know quite a bit about the effects of GS compounds in aquatic plant systems from our laboratory bioassay and small-scale outdoor barrel tests. These compounds significantly reduce main stem length in hydrilla (*Hydrilla verticillata*) and Eurasian watermilfoil (*Myriophyllum spicatum*) at parts per billion (ppb) concentrations without adversely affecting physiological parameters such as photosynthesis and respiration (Netherland and Lembi 1992). Extensive tests with one of the GS compounds, flurprimidol, showed that this compound could significantly reduce main stem length for 28 days in hydrilla at 75 ppb and Eurasian watermilfoil at 200 ppb after only a 2-hr exposure (Lembi and Chand 1992). We also developed the techniques for extracting and detecting flurprimidol in water, sediment, and submersed plant tissue using gas chromatography (Chand and Lembi 1991). Water, plant tissue, and sediment analyses showed flurprimidol half lives of 9.1,

9.9, and 178 days, respectively (Chand and Lembi 1994). Although the compound appeared to dissipate rapidly from the plant tissue, minute amounts were still present in height-reduced milfoil plants 28 days after a 2-hr treatment.

These results indicate that flurprimidol readily dissipates (probably from photodegradation) from the water but is longer lived in soil. It is taken up rapidly by the plant (maximal uptake 24 hr after exposure), and height reduction persists for at least a month and probably longer.

One of the major considerations in the use of any aquatic chemical is the route of plant uptake. Do aquatic plants take up flurprimidol from the water or from the soil or both? There are several important reasons for determining this. First, if plants can take up flurprimidol from the sediment, then it may be possible to design delivery systems that will specifically concentrate the compound in the sediment with minimal contamination of the water. Secondly, uptake from the sediment (given significant long-term persistence in the sediments) should provide long-term stem reduction in both static and flowing water systems. If, on the other hand, uptake is strictly from the water, we will probably not be able to count on significant residence time in flowing water, and delivery systems will have to be devised to extend exposure periods in hydrodynamic systems.

Our first goal this year was to initiate studies to determine the route of flurprimidol uptake. Here we describe the results of a dose-response experiment using compartmentalized plastic containers to determine whether hydrilla

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takes up flurprimidol from the soil or from the water. Our second goal was to begin a larger scale study on competitive interactions between flurprimidol-treated hydrilla and the native, beneficial species, *Vallisneria americana* (vallisneria). We know hydrilla is extremely sensitive to flurprimidol at concentrations as low as 10 ppb active ingredient (ai). Vertical stem length in hydrilla is much reduced, and the plant puts its resources into a low-growing stoloniferous form. However, little is known of the effects of flurprimidol on vallisneria. The growth form of vallisneria differs from that of hydrilla. Rather than elongating stems, most of the vallisneria height is due to leaf elongation from a short stem that remains at the ground line. If flurprimidol were to have minimal effects on leaf elongation in vallisneria, then the potential exists for significant shading of the height-reduced hydrilla by the vallisneria.

Methods and Materials

Flurprimidol uptake study

Plastic cylinders (43 cm long, 7.5 cm in diameter) were constructed with a plate to separate the water and soil compartments (Figure 1). The soil compartment was filled with a wet garden soil, and the water compartment was filled with Smart and Barko (1985) medium. A small hole in the plate allowed the hydrilla plant to be inserted into the soil. The hole was sealed with melted cocoa butter at the top of the plate and melted paraffin underneath the plate. The plants were allowed to acclimate and develop roots in the cylinders for 14 days at a light intensity of 200 $\mu\text{mol}/\text{m}^2/\text{sec}$, 25 °C, and a photoperiod of 16:8 hr light:dark. The light was reduced to 40 $\mu\text{mol}/\text{m}^2/\text{sec}$ prior to treatment. Flurprimidol applications to the water were made by introducing the compound directly into the water at the top of the cylinder. Water concentrations were 0, 1, 10, 100, and 500 ppb ai. Flurprimidol applications to the sediment were made by injecting the compound with a hypodermic needle through a small porthole into the sediment. Soil concentrations were also 0, 1, 10, 100, and 500 ppb. Each soil and water treatment consisted of

three replicate cylinders. The plants were harvested for length measurements 4 weeks after treatment. The experiment has been conducted only once to this point.

Competition study

Stock tanks (1.1 m in diameter, 60 cm in depth) were placed out-of-doors and layered with 12 cm of garden soil covered with a thin layer of sand. The remainder of the tank was filled with well water. The tanks were covered with 73 percent shade cloth. The southern form of vallisneria was planted and allowed to become established. Mean longest leaf length of vallisneria at the time of treatment was 47 cm. Hydrilla was planted 7 days



Figure 1. Plastic cylinders used in the flurprimidol uptake study. The bolts are located at the plate which separates the upper water chamber from the lower soil chamber

before treatment; mean stem length at the time of planting was 10 cm. Six plants of each species were used per tank as required. The treatments were as follows: (1) hydrilla alone, no flurprimidol; (2) hydrilla alone, with flurprimidol; (3) vallisneria alone, no flurprimidol; (4) vallisneria alone, with flurprimidol; (5) hydrilla and vallisneria mixed, no flurprimidol; and (6) hydrilla and vallisneria mixed, with flurprimidol. Each treatment consisted of three tanks. The flurprimidol concentration in the treated tanks was 150 ppb ai, applied on 3 July 1994. The plants were allowed to grow for 8 weeks at which time all plants were harvested and measured.

Results and Discussion

Flurprimidol uptake study

The selected treatments that are shown here (Figure 2) compare untreated plants with plants that were treated with the lowest flurprimidol concentration (1 ppb) used in the water and sediment compartments. Lateral stems above the plate are the laterals that were produced by the main stem in the water compartment. Laterals below the plate were actually stolons which lay on the soil surface and can get quite long, with roots produced at intervals to anchor them into the soil. The latter effect is quite typical of flurprimidol-treated plants, i.e. a switch from vertical growth to horizontal growth.

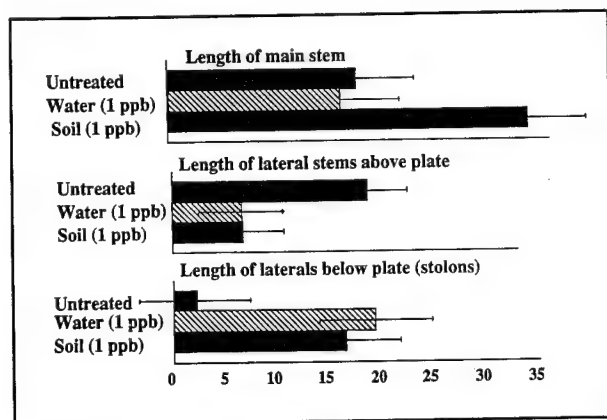


Figure 2. Effect of flurprimidol when applied to the water or soil compartments at 1 ppb

The untreated plants appeared to put their resources mostly into the production of lateral stems above the plate. Mean length of the main stem of untreated plants was 17.2 cm but mean length of lateral stems off that main stem was 19.7 cm. The mean length of lateral stems (stolons) produced below the plate (in the soil) was only 2.1 cm. The effects of the water treatment were as expected. Mean lateral stem lengths produced above the plate were considerably shorter (7 cm) than those of the untreated plants (19.7 cm). However, the lengths of stolons produced below the plate were longer than those of the untreated plants (19.4 cm versus 2.1 cm). These results suggest that flurprimidol is effective through the water even at concentrations as low as 1 ppb.

Sediment treatments mirrored water treatments in terms of length of the laterals above and below the plate (Figure 2). Main stem length was considerably longer than either the untreated or water-treated plants. This may be due to the fact that flurprimidol moves slowly from the soil system to the tip of the plant in terrestrial species. The delay in movement of flurprimidol to the top of the plant may have allowed the apical meristem to continue producing new nodes and internodes which then elongated. However, the effects on the lateral stems, both above and below the plate, even at 1 ppb, indicate that soil treatments can be as effective as water treatments.

The potential for contamination of untreated compartments was determined using gas chromatographic analysis of all soil and water samples. No flurprimidol residues were detected in the water when the soil compartment was treated; no flurprimidol residues were detected in the sediment when the water compartment was treated.

Competition study

This experiment was conducted in the summer of 1994 and will be repeated in the summer of 1995. Because of some design flaws, not all of the 1994 data are usable, but these problems should be solved when the experiment is repeated in 1995. In general, the

1994 results suggest that flurprimidol did affect leaf length of vallisneria, whether growing alone or in combination with hydrilla. However, the length of these leaves was longer than the stem lengths of hydrilla. For example, in the mixed stand tanks, the mean length of new vallisneria leaves in the untreated tanks was 52 ± 3 cm; the mean length of these leaves in the treated tanks was 35 ± 3 cm. In the treated tanks, the mean stem length of hydrilla was only 21 ± 2 cm. Of probably greater significance was the difference in shoot dry weight production between hydrilla and vallisneria in the mixed stand tanks. The mean dry weight of vallisneria in the treated tanks was 36 ± 6 g; for hydrilla mean dry weight was 4 ± 0.1 g. Visual observations of the mixed stand tanks at harvest indicated a severe reduction in the amount of hydrilla present under the vallisneria canopy, suggesting that vallisneria had a suppressive (possibly light shading) effect on hydrilla.

Flurprimidol-treated vallisneria did show marked reduction in flowering stem length. This suggests that sexual reproduction in vallisneria may well be inhibited by flurprimidol treatments; however, vallisneria rapidly propagates itself by the production of winter buds and stolons. The effects of flurprimidol on winter bud production is unknown, but our experiments indicate that although stolon length of treated vallisneria was reduced, more plants from stolons were produced in the treated than in the untreated tanks.

Future Work

Both of these experiments must be repeated in 1995. The results of the competition study are intriguing because they suggest that vallisneria may gain a competitive edge on

hydrilla when both plants are treated with flurprimidol. Since our experiment simulated the initial invasion of hydrilla into an established bed of vallisneria, other scenarios should be examined. For example, what is the effect of flurprimidol when applied to a more established bed of hydrilla and vallisneria? What is the potential for treating an established bed of hydrilla with flurprimidol and then planting vallisneria into the height-reduced hydrilla? The potential benefits of flurprimidol treatment are particularly exciting because few herbicides selectively remove hydrilla from vallisneria stands.

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Herbicide Concentration/Exposure Times: Effects on Nontarget Plants

by
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Introduction

In aquatic plant communities the introduced exotics Eurasian watermilfoil (hereafter called milfoil) and hydrilla can quickly reach nuisance levels. However, the vegetation management problems that these weeds present should not divert attention entirely from the ecologically valuable species native to the community, and target aquatics should be removed by methods that will allow beneficial plants to regenerate habitat and re-create ecosystem stability. Consequently, the most efficient use of herbicides and plant growth regulators in aquatic vegetation management requires knowledge of the effects of these chemicals on both the target and nontarget species present.

The Herbicide Concentration/Exposure Time (CET) Relationships Work Unit has produced guidelines which predict control at a wide range of aquatic herbicide dosages for milfoil and hydrilla (Green and Westerdahl 1990; Netherland, Green, and Getsinger 1991; Netherland and Getsinger 1992; Nelson, Netherland, and Getsinger 1993; Netherland, Getsinger, and Turner 1993). However, these recommendations only apply for the target species actually tested, and the Chemical Control Technology Team (CCTT) is also engaged in defining herbicide response in nontarget plants. Initial evaluations of fluridone and triclopyr effects on six nontarget species are summarized in this paper.

Materials and Methods

For these experiments rooted plants of each species were established in separate 55-L

aquaria from 40 to 60 propagules planted into a total of 10 sediment-filled 300-ml beakers, and grown in controlled environment chambers under a light intensity (PPFD) of $541 \pm 75 \mu\text{E}/\text{m}^2/\text{sec}$ 14L:10D photoperiod and water temperature of $23 \pm 2^\circ\text{C}$ (Netherland 1990). Three replications of the herbicide CETs and of untreated references were assigned to aquaria of each species in completely randomized experimental designs, and treatment concentrations were calculated on the basis of a net 47-L water volume. Pretreatment dry weight biomass data were estimated from one beaker of plants per aquaria.

Posttreatment condition of plants was evaluated visually, and various physiological parameters were monitored. Carbohydrate content, peroxidase enzyme activity, and total protein concentration were estimated using colorimetric methods (Luu and Getsinger 1990; Sprecher, Stewart, and Brazil 1993; Bradford 1976; Lowry et al. 1951). Chlorophyll concentrations were determined from dimethyl sulfoxide extractions of shoot samples (Hiscox and Israelstam 1979). Photosynthesis and respiration were determined from O_2 evolution and uptake. Phytoene and carotene pigment analyses followed those of Duke, Kenyon, and Paul (1985). Viable tissue remaining in each aquarium was harvested for final dry weight biomass measurements.

Triclopyr CET studies

Triclopyr was applied as Garlon[®] 3A, which is currently used under an Experimental Use Permit, at rates of 1 mg acid equivalent (ae)/L for 12 hr, and 2.5 mg (ae)/L (the highest labeled rate) for 24 hr. In the first of

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two separate evaluations, treatments were applied to milfoil and to the monocots sago pondweed (*Potamogeton pectinatus* L.), wildcelery (*Vallisneria americana* Michx.), and elodea (*E. canadensis* Rich.) at 5, 6, 7, and 8 weeks, respectively, after establishment from apical shoots (milfoil, elodea), winter buds (wildcelery), or tubers (sago pondweed). In the second evaluation, apical shoots of the dicot coontail (*Ceratophyllum demersum* L.), the monocot water stargrass (*Heteranthera dubia* (Jacq.) MacMill.), and the dicot watermarigold (*Bidens beckii* L.) were established and treated 2.5, 3.5, and 7.5 weeks after planting, respectively. In sago pondweed, chlorophyll concentrations were determined using three shoot samples from each aquarium at 31 days posttreatment (DAT). Peroxidase activity and total protein concentrations were determined in all species pretreatment, through 11 DAT and before final harvest.

Fluridone CET studies

This evaluation simulated an early season herbicide treatment for milfoil eradication in the Northern tier states, where fluridone applied before target weed emergence will also affect early development of the native American pondweed (*P. nodosus* Poir.), sago pondweed, water stargrass, and wildcelery. Winter buds, tubers, apical shoots, and pruned crowns of these species, respectively, were planted; after a week, emerging plants were treated with 2, 10, and 25 µg/L of fluridone. Exposure to 2 and 10 µg/L of fluridone was continued for 90 days, while the 25-µg/L treatment was removed at 60 DAT by draining

and filling aquaria twice. During the first 30 days of treatment, nominal fluridone levels at all concentrations were accidentally increased due to retreatment following flow-through periods that exchanged only a half rather than a whole aquaria water volume. Herbicide concentrations during this time are estimated to have been as shown in Table 1. Treatment levels were returned to 2, 10, and 25 µg/L at 31 days of exposure, and are discussed here as nominal levels.

Peroxidase, protein, carbohydrate, photosynthesis, respiration, and phytoene and carotene analyses were made of this material over the course of the study. Fresh weight biomass was monitored from harvest of one beaker of plants during the third, fifth, and seventh weeks after treatment, and dry weight biomass was measured on the remaining three beakers at final harvest at 90 DAT.

Results and Discussion

Triclopyr on nontargets

Evaluation of both monocot and dicot nontargets was of interest as triclopyr is an auxin-like compound, similar in herbicidal effect to 2,4-D and, in general, has activity on broad-leaf dicots rather than on monocots (WSSA 1989). The triclopyr rates evaluated here include the maximum-labeled concentration, 2.5 mg ae/L. The lower triclopyr CET has given <70 percent milfoil control in laboratory evaluations, and the higher 85 percent control (Netherland and Getsinger 1992).

Table 1
Estimated Levels of Fluridone Resulting From Retreatment of Aquaria Following Incomplete Water Exchanges

DAT	Fluridone, ug/L		
0	2	10	25
7	3	15	37.5
15	3.5	17.5	43.8
21	3.8	18.8	46.9
31	2	10	25

A range of herbicide effects was seen in target and nontarget species. In milfoil, epinastic curvature of apical and axillary shoots characteristic of auxin-like compounds was visible in all treated aquaria by 3 DAT, and epidermal rupture was evident from presence of extracellular air bubbles in stems. Stem tissue became water-logged and began to decompose, and by 14 DAT no viable stems or leaves remained. No milfoil regrowth occurred in either treatment level, and no tissue remained for biomass harvest 5 weeks after treatment. Lack of milfoil regeneration in the lower treatment rate was not expected, but removal of apical meristems for physiological analyses may have contributed to it.

In elodea, wildcelery, and coontail no decline or loss of condition was noted in plants following treatment, and differences in biomass found among treatments within these nontarget monocot species at 35 to 37 DAT were not significant (Figure 1). This is consistent with information from field treatments, including a triclopyr application made this past summer at Lake Minnetonka, MN, where coontail was unaffected while milfoil was eradicated (Getsinger 1995). Therefore, use of triclopyr to control milfoil is not expected to reduce abundance of these three species.

However, the other species in this evaluation were affected in various ways by triclopyr, and although plants were not eliminated by herbicide treatment the higher rate significantly reduced biomass in each (Figure 1). Auxin-related symptoms did not occur in the monocot sago pondweed, but by 20 DAT plants treated at both rates exhibited some loss of turgor and appeared to have less chlorophyll in leaves and shoots compared to untreated references. Cellular deterioration continued at the higher treatment level, indicated by tissue necrosis and colonization of the plant surface by algae. By 31 DAT mean chlorophyll content was $1.11 \pm (\text{s.d.}) 0.20$ and $1.12 \pm 0.37 \text{ mg (g}^{-1}\text{fw)}$ in the reference and the lower treatment aquaria, respectively, and $0.60 \pm 0.48 \text{ mg (g}^{-1}\text{)}$ in the higher treatment ($p = 0.22$), reflecting significantly reduced chlorophyll in two of three replicate aquaria. Growth continued from functioning root

crowns even at the maximum labeled rate, but at 35 DAT these pondweed plants had less than half the biomass of the lower treatment or untreated material.

Water stargrass, a monocot, was affected in a way similar to that to the pondweed, with the higher triclopyr concentration causing chlorophyll loss, necrosis, and reduced biomass, but not preventing continued growth from apices and crowns. However, this species did show the pronounced epinastic response associated with 2,4-D activity on dicots, with crooking and curling of apical tips and leaves by 1 DAT and 3 DAT in plants at the higher and lower rates, respectively, and brittle upper nodes. A return to normal shoot development with new apical growth and flowering was evident in the lower treatment by 23 DAT, but less regrowth in the higher treatment resulted in significantly less final biomass. These results indicate that in the field sago pondweed and water stargrass could be maintained during milfoil control by use of intermediate rates. At the maximum labeled rate regrowth from root crowns would be expected following herbicide application, with full revegetation in the following growing season.

Apical tips in treated material of the dicot water marigold showed growth hormone effects by 2 DAT. Reduction in internode length and epinasty caused stunting of apices and reflexive curvature of leaf petioles and stem; symptoms were more severe with the higher dose. In addition, numerous roots emerged from stem nodes in a zone just below the shoot apices in both treatments by 9 DAT. While some root formation occurred along stems of untreated plants during the course of the evaluation, in treated material the greater length (up to 18 cm) and concentration in one area of the axillary roots resembled the results of an application of auxin rooting hormone. Apical tips remained healthy in treated plants, and returned to normal growth and elongation after 15 DAT. By 32 DAT these rooted apical shoot segments were fragmenting from treated plants, and in the field could have functioned as autofragments, floating away from the parent plant to establish

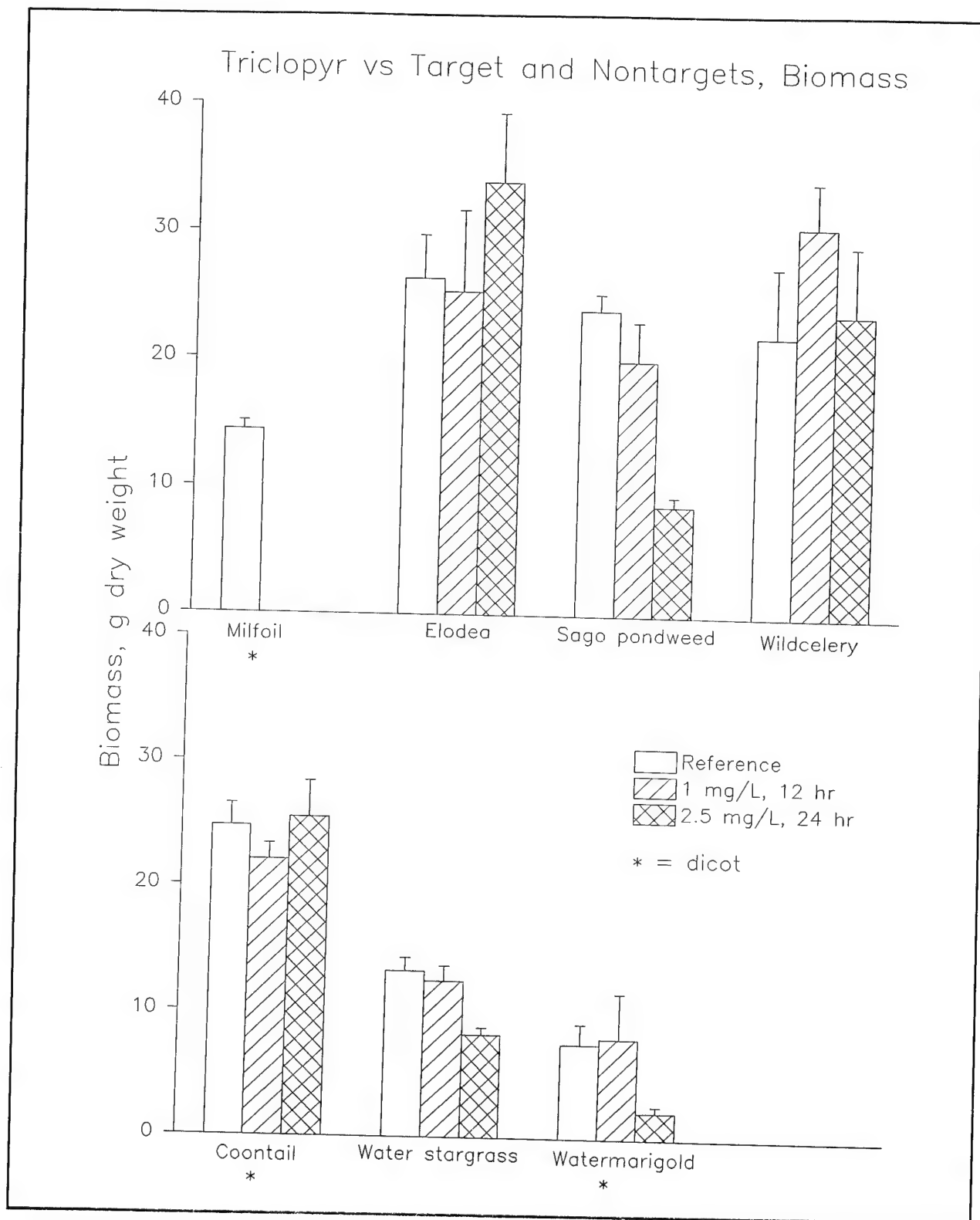


Figure 1. Final harvest (35 to 37 DAT) biomass for milfoil and six monocot and dicot nontarget species treated with triclopyr at 1.0 mg ae/L for 12 hr; 2.5 mg ae/L for 24 hr; or untreated. Bars represent the standard errors of means of three replicates

in sediment. Thus, even though overall biomass in aquaria treated at the higher rate was significantly reduced, it is possible that in the field triclopyr treatment would promote vegetative coverage of water marigold by encouraging rooting through its auxin-like properties and producing increased propagation through autofragmentation.

Physiological evaluations showed that peroxidase enzyme activity increased only with treatment in the highly susceptible dicot milfoil, while protein content and enzyme activity were not correlated to the intermediate triclopyr effects which resulted in reduced biomass (data not shown).

The varying effects of triclopyr on monocot and dicot nontarget species show that a blanket recommendation cannot be given for use of this compound as a "broadleaf herbicide" in aquatic vegetation management. However, the reduction in vigor or biomass without complete eradication seen in watermarigold, water stargrass and sago pondweed indicates the potential for regrowth and revegetation of these species following triclopyr control of milfoil.

Fluridone on nontargets

Netherland, Getsinger, and Turner (1993) have shown that milfoil and hydrilla are controlled at 12 and 24 $\mu\text{g/L}$ with 90-day exposures, and levels as low as 5 $\mu\text{g/L}$ can control hydrilla as long as exposure time can be maintained for 90 days (Netherland 1994). Results here show that the nontarget American and sago pondweeds and wildcelery tolerate a 90-day exposure to 2- to 4- $\mu\text{g/L}$ fluridone but are significantly affected by 10 $\mu\text{g/L}$ (Figure 2).

Water stargrass was initially susceptible to fluridone, showing characteristic bleaching of new growth in treated plants, but this species succumbed to a plant pathogen early in the study and did not remain in the evaluation.

The pondweeds showed no bleaching or reduction in chlorophyll with 2 $\mu\text{g/L}$, and fresh weight biomass at 4 to 5 weeks posttreatment did not differ from that of untreated material

(data not shown). American pondweed underwent a normal lifecycle at this concentration, producing surface leaves, flowers, and seed. By 90 DAT it retained significantly more biomass than untreated material which was further advanced in senescence. Sago pondweed was slowed in biomass accumulation by 2 $\mu\text{g/L}$, but continued new growth through 90 DAT. Fluridone at 10 and 25 $\mu\text{g/L}$ eliminated almost all tissue in both pondweeds by 60 DAT, with no appreciable regrowth in the 30 days following removal of the 25- $\mu\text{g/L}$ treatment.

Growth from pruned mature crowns of wildcelery was uneven (see pretreatment biomass, Figure 2) and much slower than seen normally from winter buds. Fluridone at 2 $\mu\text{g/L}$ showed no bleaching effect or growth reduction relative to untreated plants, but biomass was reduced by the two higher treatments.

The various physiological analyses reflected fluridone effects during the first weeks of treatment, but later loss of tissue prevented further conclusions from most of these measurements (data not shown). Standing plant populations of these species are expected to be adversely affected by fluridone control of target weeds; however, the vegetative reproductive structures of these species (winter buds, stolons, crowns, tubers) may be able to renew plant populations following fluridone dissipation. Hydrilla turions and tubers are reduced in number by 5- $\mu\text{g/L}$ fluridone (McDonald et al. 1993), and suppression should be investigated in these nontarget species which rely on underground propagules.

Future Work

Future studies will monitor additional treatment levels to determine minimum threshold concentrations of triclopyr and fluridone versus these and other nontarget species that are prominent in the Southern and Northern plant communities. Evaluations of treatment effects on vegetative propagules will indicate the long-term regrowth and revegetation response that can be expected in the field. This information will increase the predictability of herbicide effect on valuable native species in the field, and allow herbicide applications to be

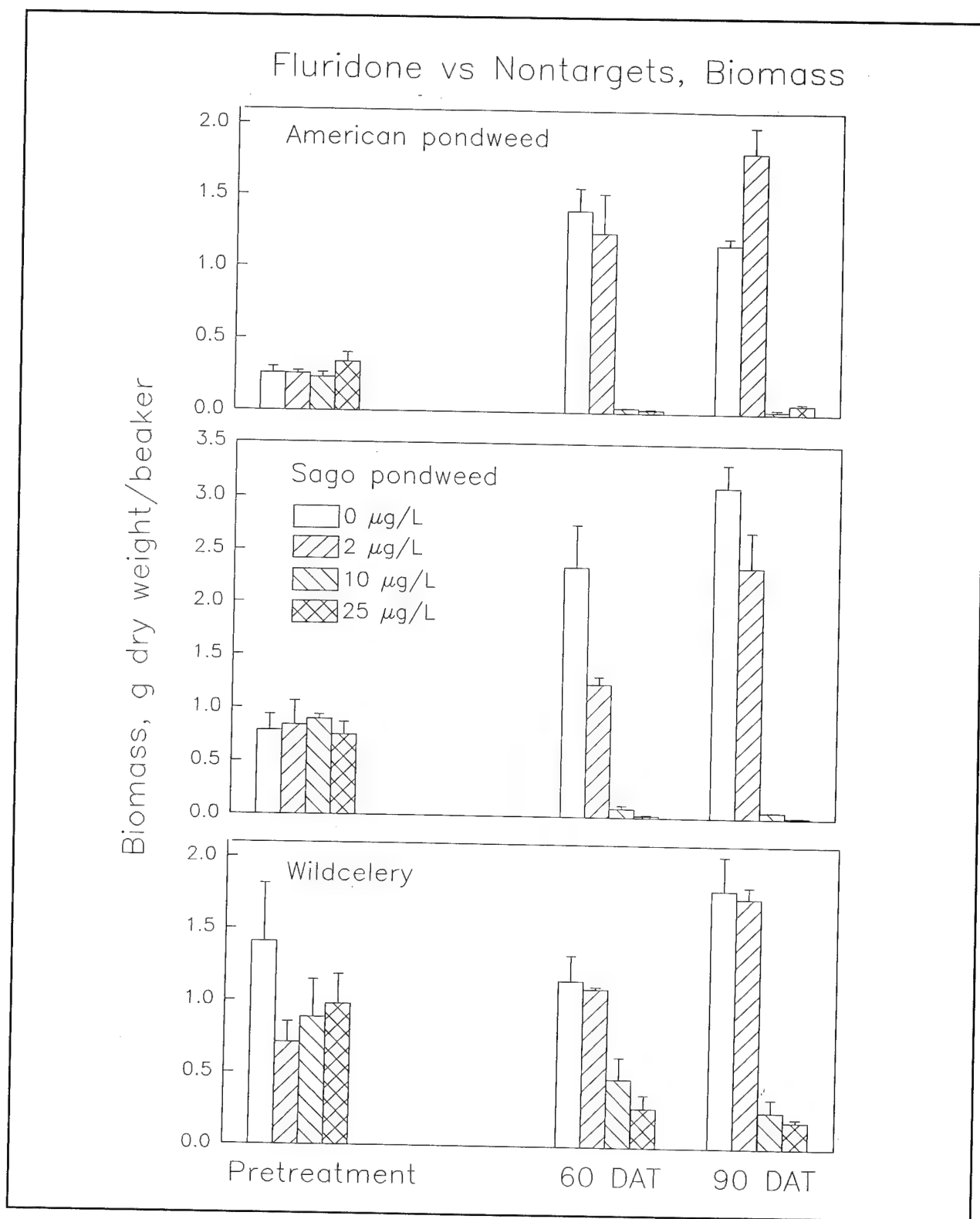


Figure 2. Pretreatment, 60, and 90 DAT biomass (one planting beaker) for three nontarget species treated with fluridone at 2, 10, and 25 $\mu\text{g ai/L}$ for 90 days, or untreated. Bars represent the standard errors of means of three replicates

tailored precisely to the site and the plant community present. The details of nontarget species' response to various concentration and exposure times can provide basic information to the CCTT's work units dealing with selectivity and integrated control using herbicides in conjunction with weed pathogens. These approaches increase the potential of herbicides for eliminating target plants while benefiting individual native species and the entire plant community during vegetation management.

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Mesocosm Evaluation of a New Endothall Polymer Formulation

by
Michael D. Netherland¹ and E. Glenn Turner²

Introduction

The conditions under which an aquatic herbicide is applied (high or low water exchange, thermal stratification, water quality and temperature, plant species and density) will have a direct influence on field performance (Fox et al. 1991). The formulation of an aquatic herbicide (liquid, granular, slow-release pellet) will also affect field performance (i.e., distribution, concentration, availability, and efficacy) under diverse environmental conditions (Getsinger, Haller, and Fox 1990). In particular, properties of granular formulations (carrier type (clay, lignin, gypsum, polymer), active ingredient (ai) loading, and release rates) can greatly affect aqueous behavior. Therefore, development of new granular formulations requires testing at the laboratory and mesocosm scale to determine suitability for large-scale field testing.

In addition to performance characteristics, herbicide formulations can also impact the applicator. For example, Aquathol[®] (10.1 percent dipotassium endothall granule) has demonstrated excellent field performance; however, applicators have complained of the high bulk required for treatment (90 percent inert) and the tendency of the granules to form an irritating dust. In response, the manufacturer of endothall (Elf Atochem NA) has begun development of a new superabsorbent polymer formulation (SPF) that contains dipotassium endothall at 63 percent ai with a very low potential for dust formation. Although the SPF is promising it remains untested. Primary research needs include determining efficacy versus conventional formulations, release and distribution characteristics, and handling char-

acteristics at the laboratory and mesocosm level prior to committing to full-scale field testing.

The potential advantages of reducing the bulk of the SPF are clear. For example, an endothall treatment (3.0 mg/L) of a 2-hectare area 2 m in depth would currently require 1,095 kg of Aquathol[®] (10.1 percent ai), whereas the SPF (63 percent ai) would require only 185 kg of material. Reducing the bulk of the SPF could decrease application time (less reliance on nurse boats or onshore loading), reduce handling and exposure (a 200-lb granular hopper would only need to be filled twice versus 10 times for the conventional formulation). Moreover, the SPF will result in significantly lower dust formation.

The SPF is composed of crosslinked polyacrylate/polyacrylamide polymers, which are currently used in agricultural and greenhouse applications (Stockosorb[®] 400 series) to improve soil moisture retention and uniformity. Upon contact with water, the granules absorb water and expand (releasing endothall) to form a gel-like material. It is important to note that the polymers (minus the ai endothall) are recognized as nontoxic and nonhazardous (Stockhausen 1985). Development of inert nontoxic formulations is a key objective in this work unit, as this ensures environmental compatibility and a greater likelihood of approval for use by the EPA.

The objectives of this work were to evaluate the laboratory release profiles of the SPF versus the conventional granule and to compare efficacy, aqueous distribution, and handling characteristics of the SPF versus the liquid and conventional granule formulations

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at the mesocosm/field level. In addition, one metered treatment was employed to continue work designed to deliver threshold levels of endothall over an extended exposure period.

Materials and Methods

Laboratory release profiles were determined by placing the SPF and Aquathol^R granules in 52-L aquaria to achieve a total concentration of 6.0 mg/L. A general water culture solution proposed by Smart and Barko (1984) for growing submersed macrophytes was used for these studies. Water was gently bubbled to provide limited mixing, and water temperature was maintained at 22 ± 1 °C throughout the study. Water samples were collected (middepth) at 0, 0.5, 2, 12, and 24 hr posttreatment. At 24 hr all aquaria were drained and refilled three times (no granules were removed) and water was sampled on day 2 at 2, 12, and 24 hr. This procedure was repeated for day 3. Treatments were replicated four times. Following collection, samples were immediately frozen and shipped to A&S Environmental (Lancaster, PA) for analysis.

The mesocosm/field scale studies were conducted at the TVA hydraulic flume system (located at the TVA Aquatic Research Laboratory, Browns Ferry, AL) previously described by Turner, Netherland, and Burns (1992) and Turner et al. (1993). Each of the eight flumes used for this study measured 112 m in length, 4.3 m in depth, and were lined with a 50-cm-thick layer of reservoir sediment. Water depths (1.2 m) were maintained by stacking weir boards at the outlet end of each flume. Water discharge rates measured at the outlet ranged from 65 to 75 m³/hr or 3.2 to 3.7 volume exchanges (flume volume is calculated at 550,000 L) per day.

Flumes were drained in early May and given a 3-week drying period prior to planting Eurasian watermilfoil (hereafter called milfoil). Two stands of milfoil (4.3 by 12 m) were planted at the middle and outlet end of each flume. Water flow was restored and maintained throughout the rest of the study. Milfoil was given a 2-month period for establishment. In addition to milfoil, species such as *Najas*

sp., *Potamogeton* sp., *Ceratophyllum*, and *Chara* germinated and represented the majority of plant biomass in each flume. In fact, in several flumes poor milfoil establishment was attributed to aggressive competition from these species. Several flumes had nearly 100 percent surface cover prior to herbicide treatment. Pretreatment biomass samples were randomly collected with 0.5-m² PVC frames at four sites within each plant stand.

Flumes were treated with either 3.0 mg/L of liquid endothall (Aquathol K^R), granular endothall (Aquathol^R), SPF (two flumes), or 1.5 mg/L of SPF (two flumes). In addition, one flume was treated using a metering pump calibrated to deliver 0.4 mg/L for either 48 hr (plant stand 1) or 72 hr (plant stand 2). Both the metered and liquid treatments included the use of rhodamine WT dye to allow on-site, real time confirmation of likely endothall residues. One flume was also established as an untreated control.

Following treatment, water samples were collected at middepth from flumes at 0, 0.5, 2, 6, 12, 24, 36, 48, and 72 hr posttreatment. Furthermore, flumes treated with SPF were sampled at three depths to determine distribution through the water column. Water samples were immediately frozen and sent to Elf Atochem (Bryan, TX) for residue analysis. Water flow was maintained throughout the treatment and measured daily at the metal weirs. As of the writing of this update, water residues analysis had not been completed and will therefore not be reported in this paper.

Visual observations of treatment effects were noted on a weekly basis and biomass was collected at 8 weeks posttreatment. The same procedure was used as described for pretreatment biomass; however, sampling at the post-treatment harvest was more intensive than pretreatment sampling.

Results and Discussion

In the laboratory studies the SPF granules sank to the bottom of the water column upon treatment and within minutes a noticeable increase in granule size (due to absorption of

water) was observed. By 1-hr posttreatment the granules (2-3 mm) had increased in size two- to three-fold and had also changed from a hard pellet to a clear gelatinous cube. Results of laboratory release rate studies indicated that both the conventional granule and SPF rapidly released endothall with >80 percent released from the granules within 2 hr (Table 1). Release from the SPF was slightly reduced compared to the conventional formulation. In contrast to the conventional granule, some endothall was released from the SPF on day 2. Although statistically significant differences were noted in release rates, these differences were minimal and unlikely to negatively impact field performance. Moreover, the ability to extend release rates from the SPF may have potential applications for controlled-release technology.

Table 1
Endothall Laboratory Release Data
from the Conventional 10.1% Clay
Granule (Aquathol^R) and the SPF

Time hr	Aquathol ^R % released	SPF % released
0.5	85	67
2	93	85
12	99	92
24	100	96
48	—	100

Results of the hydraulic flume studies showed that all endothall treatments significantly reduced plant biomass at 8 weeks posttreatment. Although the liquid endothall treatment of 3.0 mg/L resulted in an increase in milfoil biomass in plant stand 3 (low exposure time), an 88 to 90 percent reduction in *Najas* sp. biomass was noted in plant stands 2 and 5 (Figure 1). Milfoil was planted in stand 6, but was outcompeted by *Najas* sp. to the extent that no milfoil was detected in pre- or posttreatment harvests. The Aquathol K granule and SPF at 3.0 mg/L resulted in similar control of both milfoil and *Najas* sp (Figures 1 and 2). The SPF provided >90 percent control of both milfoil and *Najas* sp. while the

Aquathol K granule gave >80 percent control of both milfoil and *Najas* sp. The SPF endothall rate of 1.5 mg/L yielded 76 to 93 percent milfoil control and >90 percent control of *Chara*, *Najas*, and *Ceratophyllum* (Figure 2). Little difference was noted between the 3.0- and 1.5-mg/L SP treatments. All formulations tested provided season-long control of submersed vegetation in high-flow environments.

The metered application of 0.4 mg/L for 48 and 72 hr resulted in a significant overall decrease in milfoil biomass (70 to 80 percent) and *P. crispus/Najas* sp. biomass (80 to 96 percent) (Figure 3); however, milfoil control achieved at 48 hr of exposure was greater than control achieved at 72 hr. This result was unexpected as there is generally a direct relationship between increased plant control and increased exposure time with endothall (Netherland, Green, and Getsinger 1991). Previous flume studies showed that endothall at 0.5 mg/L for 96 hr resulted in 100 percent milfoil control (Netherland et al. 1994). These results (in combination with limited laboratory data) suggest that 0.3 to 0.4 mg/L of endothall may represent a threshold for submersed plant control.

Milfoil in the untreated control flume was completely eliminated at 8 weeks posttreatment (Figure 3). In this case, the disappearance of milfoil in plant stands 2 and 5 was attributed to very aggressive competition from *Potamogeton nodosus*. The floating pondweed leaves formed a complete canopy at the surface, and it is likely that as it continued to expand the established pondweed preempted milfoil growth from newly planted apical tips. The control flume was the only flume with significant stands of *P. nodosus*.

Summary

Results from these studies indicate that the SPF provides a very similar release profile to the current conventional granule. Furthermore, large mesocosm-scale efficacy tests showed the SPF to be as or more effective than the current conventional granule and liquid products. Aside from the formulations,

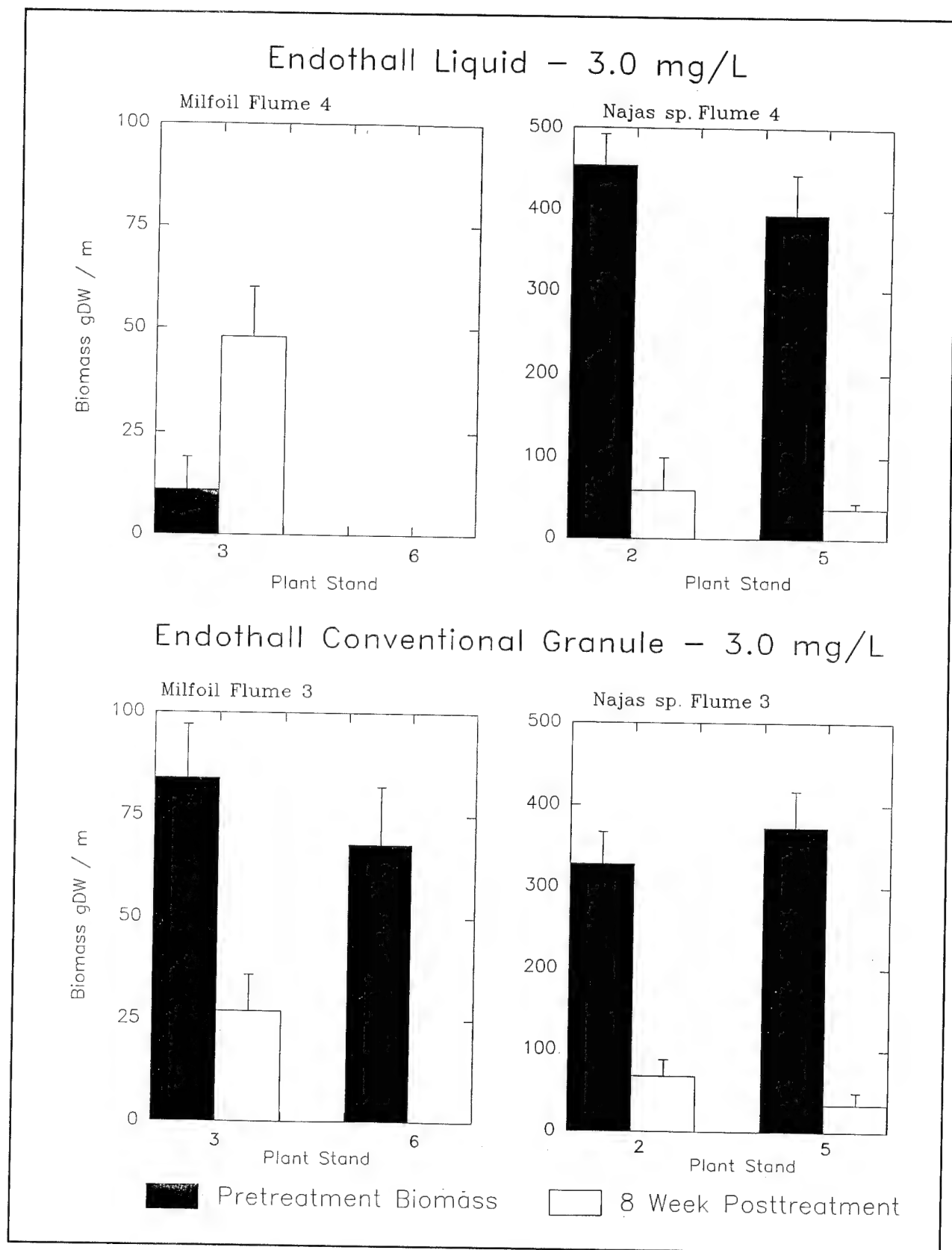


Figure 1. Endothall efficacy on Eurasian watermilfoil and *Najas* sp. following a 3.0-mg/L treatment to flowing water flumes with the conventional liquid or conventional granular formulation. Each bar represents the average of four biomass samples taken from each plant stand

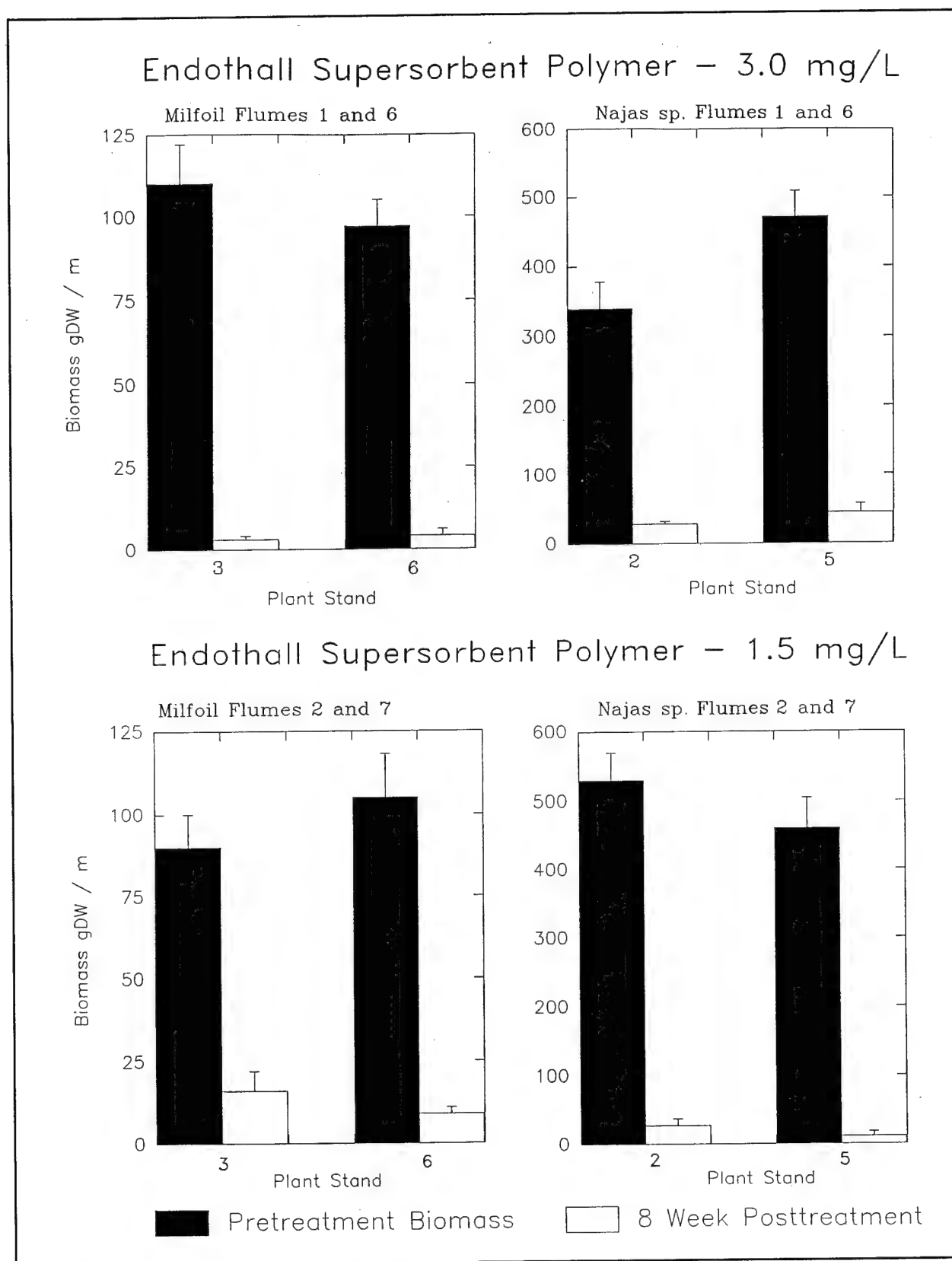


Figure 2. Endothall efficacy on Eurasian watermilfoil and *Najas* sp. following a 1.5- or 3.0-mg/L treatment to flowing water flumes with a supersorbent polymer formulation. Each bar represents the average of four biomass samples taken from each plant stand

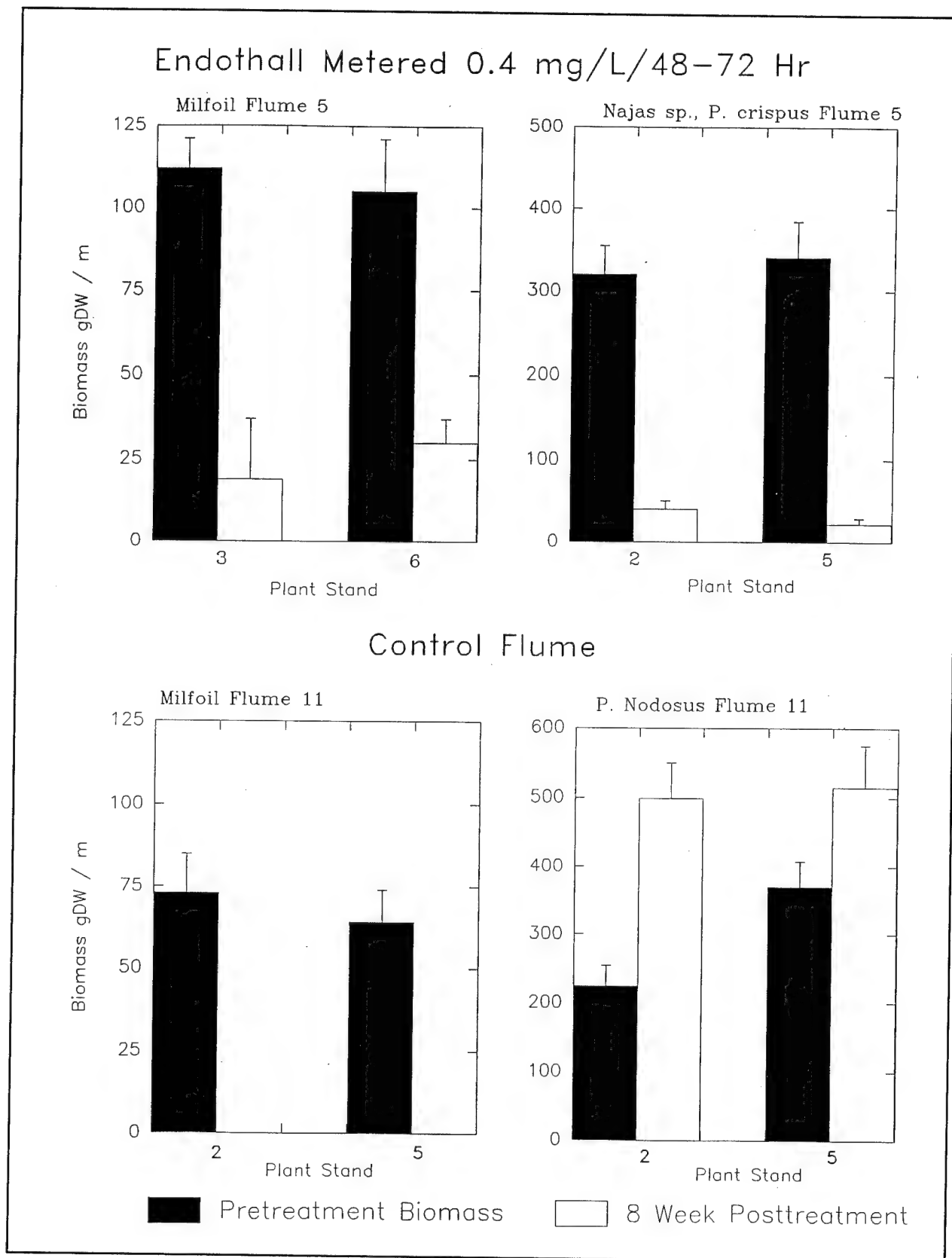


Figure 3. Endothall efficacy on Eurasian watermilfoil and *Najas* sp. and *P. crispus* following a metered treatment of 0.4 mg/L to flowing water flumes or untreated control flume. Each bar represents the average of four biomass samples taken from each plant stand

the major differences between the conventional and the SPF granule are the active ingredient loading rate (10.1 percent for the conventional granule and 63 percent for the SPF granule) and the reduced potential for dusting with the SPF. Although the SPF product did not greatly increase efficacy, the potential for a significant improvement in handling and reduced applicator exposure justify continued research with this product.

Future Work

Plans for future work include large-scale field dissipation and efficacy studies with the SPF granule. These studies will be conducted in waterbodies in Florida in 2- to 4-hectare plots. Efficacy and dissipation will be compared between the conventional granule and the SPF. In addition, handling characteristics and dust formation will also be compared between the two products. Obtaining this information will be crucial for a data package submission to EPA for consideration of registration of this product. In addition, water residues collected from the flume studies will be analyzed to determine aqueous distribution and release rates at the mesocosm level.

Other work will include continued development of controlled-release or metering technology to deliver threshold concentrations of herbicide over an extended exposure period (48-120 hr). A large-scale demonstration research project using one of these technologies is planned for irrigation canals in the Western states in FY95.

Acknowledgments

The authors would like to thank Michael Bull, Charles Mayfield, Carl Wilmer, and Tommy Woods for technical assistance and Dan Haraway and Jennifer Moses of the TVA Aquatic Research Lab for logistical support. The cooperation of Elf Atochem, North America, for providing endothall formulations and residue analyses is greatly appreciated.

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Reduced Rate Endothall Application for Controlling Sago Pondweed in High-Flow Environments

by

David Sisneros¹ and E. Glenn Turner²

Introduction

Recent studies at the TVA hydraulic flumes indicate that providing continuous exposure (48-96 hr) to low herbicide concentrations can be as or more efficacious than traditional application at higher label rates. Moreover, the use of low herbicide rates (near the potable water tolerance) can often decrease toxicity to nontarget organisms and can potentially reduce restrictions on water use. In flowing-water systems traditional slug applications of certain herbicides may not work due to rapidly dissipating concentrations and a concomitant lack of exposure period (Hansen, Oliver, and Otto 1983). This study was designed to evaluate the efficacy of metering low concentrations (10 times below the maximum label rate) of the herbicide endothall for control of *Potamogeton pectinatus* L. (sago pondweed) in the high-flow environment of irrigation canals.

Herbicide concentration/exposure time requirements for various herbicides have been studied by the WES (Green and Westerdahl 1990; Netherland, Green, and Getsinger 1991; Netherland and Getsinger 1992; Netherland, Getsinger, and Turner 1993). Numerous herbicides have been evaluated in laboratory studies demonstrating that extended exposure to low concentrations can provide good to excellent efficacy. This concept was evaluated further by the WES and the U.S. Bureau of Reclamation (BOR) (Netherland and Sisneros 1993) in large hydraulic flume studies in Alabama in 1993. In these studies controlled-release gypsum matrices and metering pumps were used to deliver threshold concentrations of triclopyr to control Eurasian watermilfoil.

During the 1993 study, limited testing of the herbicide endothall was conducted using the controlled-release gypsum technology in flowing water to determine efficacy on aquatic vegetation (Eurasian watermilfoil, *Najas* sp., *Potamogeton* sp.) (Netherland et al. 1994). Approximately 0.5 mg/L of endothall released for 96 hr resulted in complete control of aquatic vegetation throughout the flume. From this study it was determined that the broad spectrum of control provided by endothall and the potential to greatly reduce use rates (while maintaining good efficacy) made it an excellent candidate for testing in the high-flow environment of irrigation canals choked by aquatic vegetation.

Materials and Methods

A field site was selected in the BOR Pacific Northwest Region (Figure 1), in the Minodoka Project, on land operated and maintained by the North Side Canal Company at Jerome, ID. The S19 canal at this study site (Figure 2) was approximately 25 miles west of Twin Falls, ID. The portion of the S19 used was earthen and approximately 14 ft wide and 2 ft deep. The study site was approximately 3 miles long. Flows in the system ranged from 12 to 18 cfs and consisted of return flow water from agricultural irrigation. The length of the canal to be treated was 2 miles. All treated water entered a 2- to 3-acre augmentation pond where diminishing residues were further diluted with inflow water from another irrigation canal. All water leaving the augmentation pond was diverted through a canal and used for power production before entering the Snake River.

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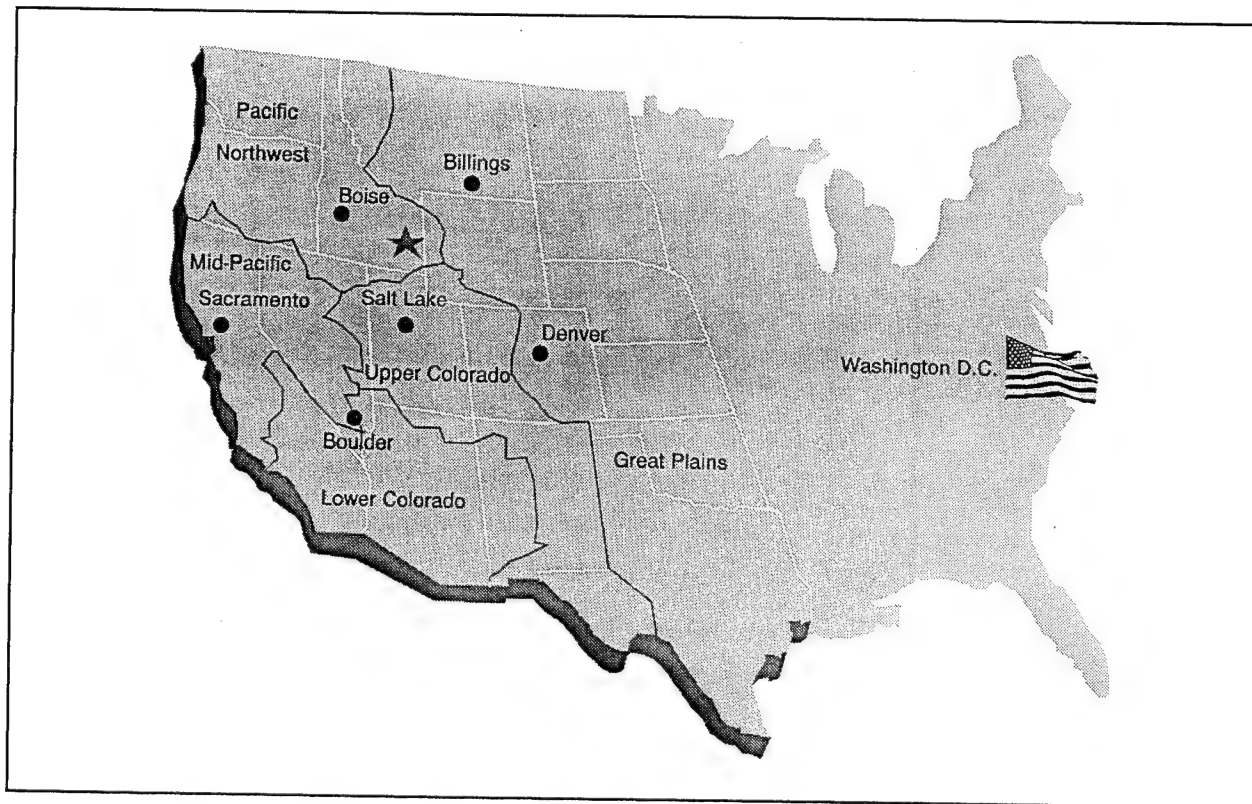


Figure 1. Location of field site (within Minodoka Project, Pacific Northwest Region of U.S. Bureau of Reclamation)

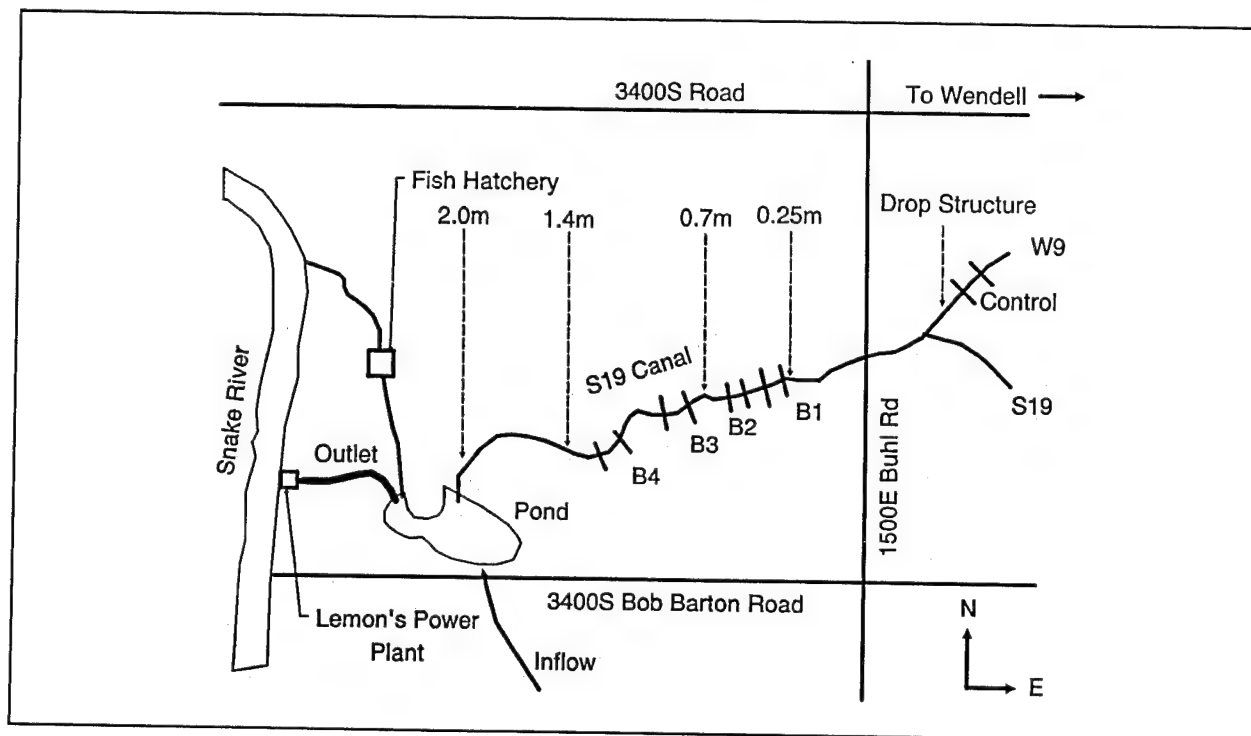


Figure 2. Location of the S19 canal showing the location of sampling stations at 0.25, 0.7, 1.4, and 2.0 miles from the drop structure where endothall was metered in. Additional sampling stations included the fish hatchery and the outlet at Lemon's Power Plant

There were five biomass sampling sites within the study area to determine herbicide efficacy. Each biomass site was 100 ft long with the control biomass site located just above the application site. Two of the four biomass sites were established dependent on plant density (visual observations) between the application site (drop structure) and the 0.7-mile sampling site. The remaining two biomass sites were randomly located between the 0.7-mile sampling site and the 1.4-mile sampling site. No further biomass sites were established downstream of the 1.4-mile sampling site due to decreasing water velocities and increasing water depths created by the influence of the small holding pond.

Six random biomass samples were taken from each of the biomass sampling sites using a 0.25-m biomass square prior to treatment and at 21 and 41 days posttreatment. Biomass samples were then sorted to species and dried for 24 hr at 100 °C to determine dry weight/square meter. The major aquatic weed species was *Potamogeton pectinatus* L. (sago pondweed) while the minor aquatic weed species was *Alisma* L. (water plantain).

Seven water sample sites were established for endothall residues at 0.25, 0.7, 1.4, and 2.0 miles downstream of the application site. Three additional sampling sites were established downstream of the pond. Duplicate water samples were collected from the seven sampling sites at pretreatment and 12, 24, 36, 48, 60, 72, 84, and 90 hr posttreatment. Water samples were frozen following collection and stored for analysis.

The potassium salt of endothall (Aquathol K) was applied to the S19 canal using an electronic metering pump manufactured by Liquid Metronics Division of Milton Roy. The calculated target rate to be achieved was 0.4 mg/L for 72 hr. In addition rhodamine WT dye was added to concentrated herbicide to provide a dye concentration of 4 µg/L. Real-time readings of dye concentration on a fluorometer allowed onsite assessment of the general accuracy of the metered application (Turner, Getsinger, and Netherland 1994).

Results

Due to the persistence of drought and unanticipated irrigation requirements in the study area it was difficult for the North Side Canal Company to maintain a constant flow for this study. Preliminary residue data from the sampling site at the 0.7-mile sampling site over 72 hr indicated that the target rate of 0.4 mg/L of endothall was not achieved with the exception of the 48-hr sampling interval (Figure 3). Endothall residues at the Outlet Lemon's Power Plant (OLPP) below the pond were less than 0.10 mg/L and, therefore, never exceeded the potable water tolerance of 0.20 mg/L.

Data from the four sampling stations showed that the biomass was reduced by 38 to 53 percent at 21 days posttreatment and 30 to 70 percent at 41 days posttreatment (Figure 4). However, both sago pondweed and water plantain harvested from the treated plots showed significant necrosis and discoloration of leaves, stems, and roots. In addition, treated plants easily pulled away from the sediment and were observed to have lost buoyancy from one-week posttreatment until the 41-day posttreatment sampling. Plants harvested from the control site maintained a surface canopy with viable healthy roots which did not easily pull from the sediment throughout the study.

Conclusions

Although the target rate was generally not achieved due to variable flow rates in the canal, utilization of metering pumps with the ability to respond to fluctuations in the flow rate could overcome this problem. Visual differences between treated and control plants were dramatic. Had the target treatment concentration been achieved or the exposure period extended to 96 hr, it is likely biomass reduction would have been significant. Although quantitative biomass numbers did not show significant differences, loss of buoyancy and general necrosis of the treated plants resulted in less resistance to water flow through the system. Project managers were generally

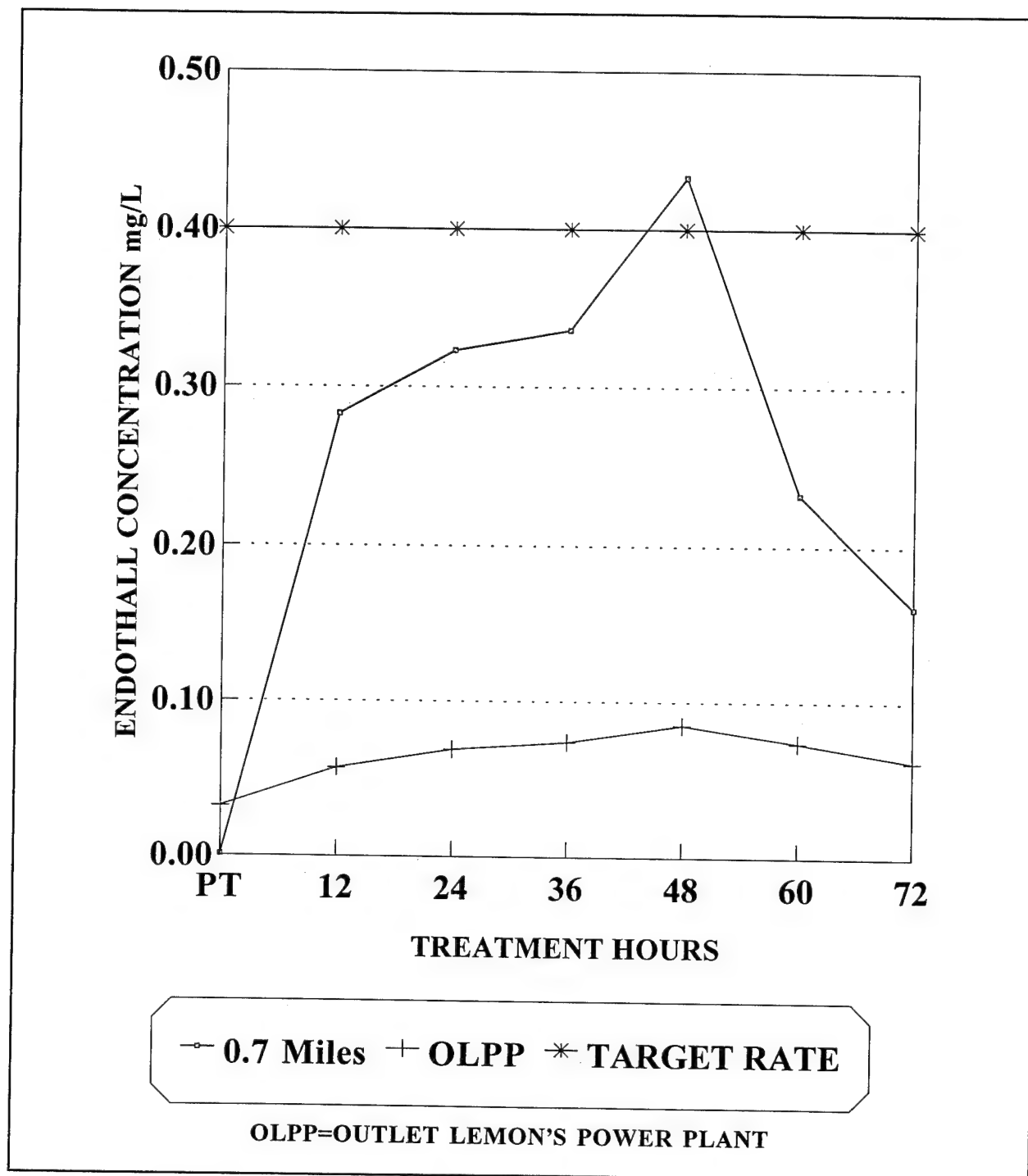


Figure 3. Endothall concentrations measured at 0.7-mile marker and the outlet at Lemon's Power Plant

pleased with the treatment and the increased flow rates due to the lack of vegetation through the water column. In addition to reducing the problem of aquatic vegetation impeding water flow, the following environmental concerns were met: (1) The potable water tolerance of 0.2 mg/L was not exceeded in the discharge

water, (2) Although treated water was not used for irrigating crops, the extremely low residues (0.10 mg/L) would likely be of very low toxicity to any crops irrigated with this water, and (3) residues were at levels which would not be detrimental to nontarget organisms such as fish and aquatic invertebrates.

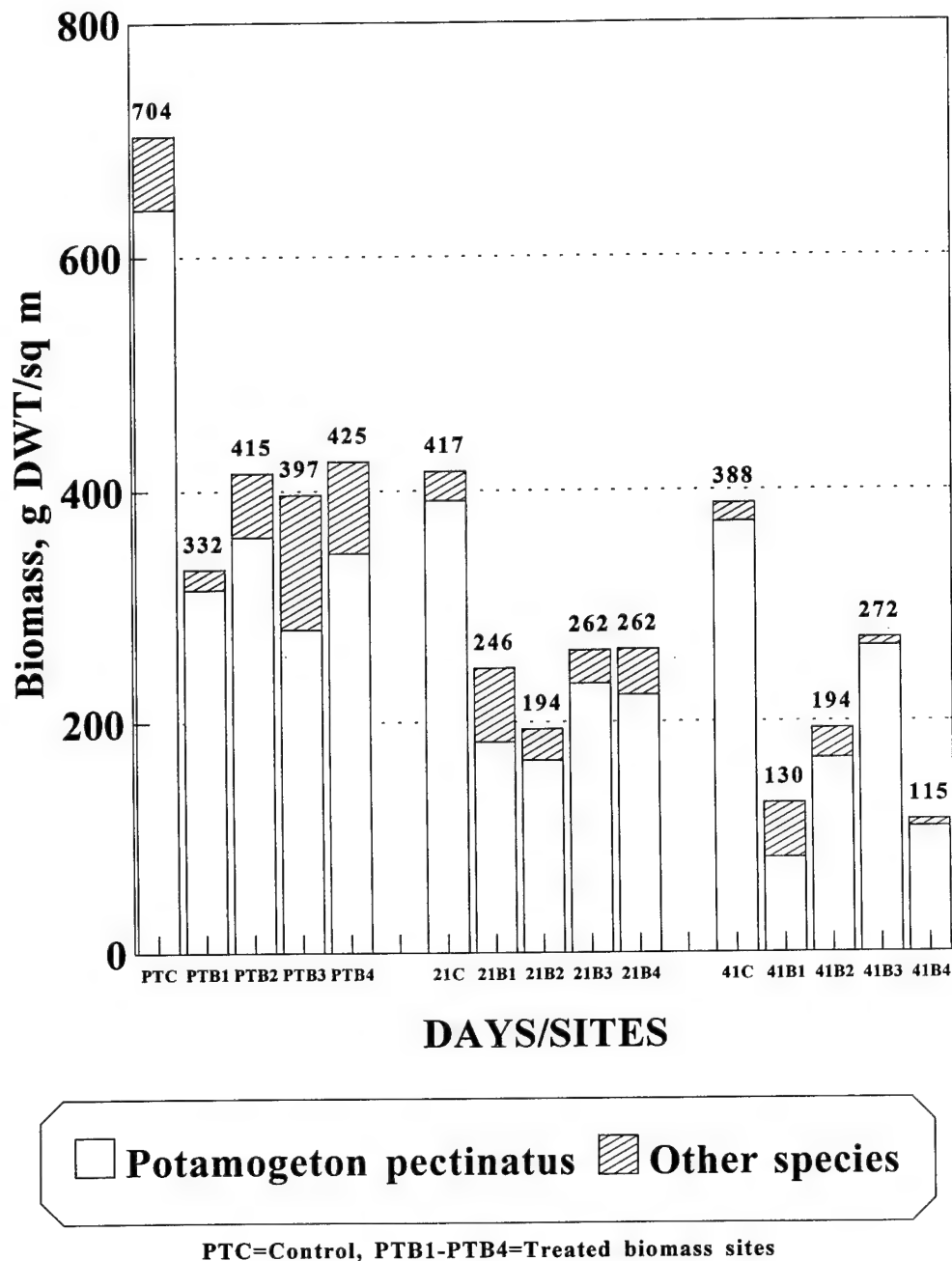


Figure 4. Biomass of aquatic vegetation from five sampling sites (control, B1, B2, B3, and B4) at pretreatment (PT) and 21 and 42 days posttreatment

Future Work

Further field tests of metering and controlled-release delivery systems are required to evaluate the utility and feasibility of using this technology (low herbicide rates for an extended exposure period) for irrigation canals

and transferring the technology to riverine and reservoir systems. These tests will need to be conducted under varying flow regimes, plant species and density, and target treatment areas. Further field testing of endothall utilizing metering pumps is anticipated in western irrigation systems during 1995.

Acknowledgment

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Preliminary Report on Species-Selective Control of Eurasian Watermilfoil with Triclopyr

by

R. Michael Smart,¹ Kurt D. Getsinger,² Gary O. Dick,¹ and John G. Skogerboe¹

Introduction

Exotic weedy species such as Eurasian watermilfoil (*Myriophyllum spicatum*) cause problems in many Corps of Engineers reservoirs and waterways. These problem species exhibit high rates of growth and have the potential to spread rapidly throughout unvegetated or disturbed systems (Smart and Doyle, in press). The prolific growth of these species produces high levels of plant biomass and a dense canopy of intertwined, leafy shoots at the water surface. This dense canopy inhibits the exchange of gases across the air-water interface and impedes wind-driven mixing of the water column, often contributing to problems with oxygen depletion (Honnell, Madsen, and Smart 1993). The dense surface mat of leaves also prevents sunlight from penetrating the water column, shading out seedlings of native plants that might have been potential competitors. Once it becomes well established in disturbed plant communities, Eurasian watermilfoil can even displace existing native plant species (Madsen et al. 1991).

As a result of its weedy growth characteristics, Eurasian watermilfoil often grows in large, monospecific beds. The low diversity of plants in these Eurasian watermilfoil-dominated environments, coupled with the poor water quality conditions that can occur under the surface canopy (Honnell, Madsen, and Smart 1993), result in poor-quality, low-diversity aquatic habitats.

Most native plant species do not cause problems with water resources, but do provide valuable aquatic habitat for fish, water-

fowl, and other aquatic life, as well as stabilizing sediments and improving water clarity and quality (Smart et al., in press). One goal of management should be to selectively control the growth of Eurasian watermilfoil and restore diverse native plant communities. These native plants would provide ecological benefits while occupying the niche vacated by the exotic weedy species, thereby preventing or slowing reinvasion (Smart and Doyle, in press).

Materials and Methods

The experiment was conducted in the Chemical Control Mesocosm System located at the Lewisville Aquatic Ecosystem Research Facility in Lewisville, TX. The experiment employed 17 large fiberglass tanks supplied with filtered lake water from a lined water-supply pond. Detailed characteristics of the system have been provided elsewhere (Dick, Getsinger, and Smart 1993; Smart and Decell 1994).

In October 1993, Eurasian watermilfoil, elodea (*Elodea canadensis*), and water star grass (*Heteranthera dubia*) were individually planted in 4.4-L plastic containers of sterilized pond sediment. These, and additional unplanted containers, were held in a growout pond throughout the fall and winter period. In early March 1994, planted and unplanted containers were moved to the experimental tanks, and the unplanted containers were planted with vallisneria (*Vallisneria americana*) or sago pondweed (*Potamogeton pectinatus*). Each tank held 54 containers of plants (33 containers of Eurasian watermilfoil and 5 containers of

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each of the four native plant species (Figure 1). The greater number of containers of Eurasian watermilfoil ensured that the experimental tank communities would be dominated by Eurasian watermilfoil. Plants were allowed to acclimate for several weeks prior to treatment.

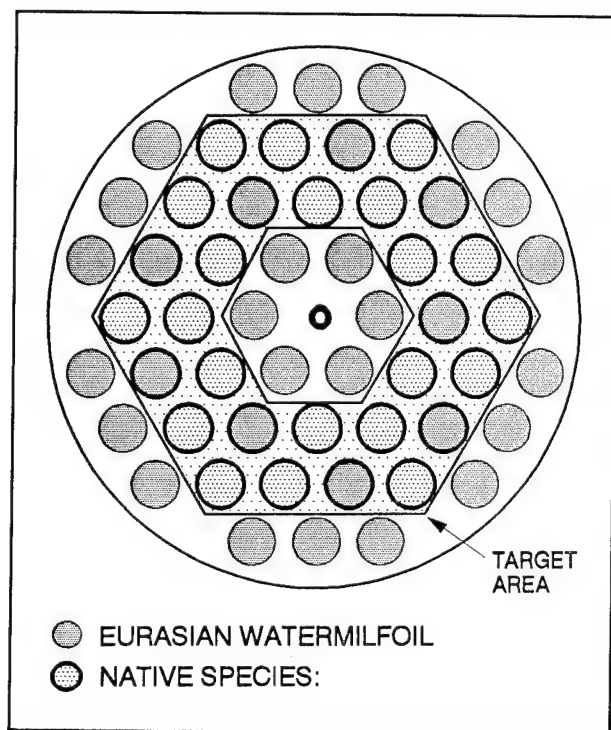


Figure 1. Arrangement of containers in experimental tank communities. Native plant species were randomly assigned to positions within the indicated target area. Only Eurasian watermilfoil containers located within the target area were sampled for biomass

Triclopyr was applied to the tanks in early April 1994 to give concentrations of 0.0, 0.5, or 1.0 mg/L. Different exposure times were achieved by varying the rate of flow of water supplied to the tanks. Flow rates were calibrated to provide half-lives of 6, 12, or 24 hr. Three replicate tanks of each of five treatments were used: 0.0 mg/L (reference), 0.5 mg/L \times 12 hr, 0.5 mg/L \times 24 hr, 1.0 mg/L \times 6 hr, and 1.0 mg/L \times 12 hr. A single tank was treated at maximum label rate (2.5 mg/L) under static conditions (no flow) to evaluate the maximum potential impact of a triclopyr application on the selected native plants. Regular visual observations indicated that the

1.0 mg/L \times 6 hr treatment was ineffective at controlling Eurasian watermilfoil, and these tanks were retreated in early May 1994, with 1.0 mg/L \times 24 hr. Rhodamine WT dye was added to each of the tanks as a tracer of the herbicide and to verify dilution rates.

Pretreatment biomass was estimated by harvesting nine replicate Eurasian watermilfoil and five replicate containers of each of the native species from a single representative tank that had been established for that purpose. Plant shoots from each harvested container were cut at the sediment surface, rinsed in filtered lake water, separated to species, and dried at 60 °C to constant weight. At termination of the experiment in late July 1994, final biomass was determined in the same manner.

Results

Biomass of Eurasian watermilfoil was significantly reduced at all but the lowest exposure (Figure 2). Effective control of Eurasian watermilfoil required exposure to 0.5 mg/L for greater than a 12-hr half-life or exposure to 1.0 mg/L for greater than a 6-hr half-life. As little as one fifth of the label rate, applied under conditions providing a half-life of only 24 hr, markedly reduced the growth of this exotic species. Eurasian watermilfoil was completely eliminated from the tank treated with the maximum label rate.

Total community biomass was not greatly affected by any of the treatments (Figure 3). Although biomass of Eurasian watermilfoil was reduced under all but the lowest exposure to triclopyr, biomass of the native species increased, maintaining total community biomass at roughly similar levels across all of the replicated treatments. Although final biomass of Eurasian watermilfoil was not reduced at the lowest exposure (0.5 mg/L \times 12 hr), its growth was sufficiently arrested to greatly reduce its suppression of the native species. Community biomass attained its maximum level in this treatment. Although the maximum label rate treatment was unreplicated, native species biomass attained in this treatment appears to be somewhat reduced relative to the lowest

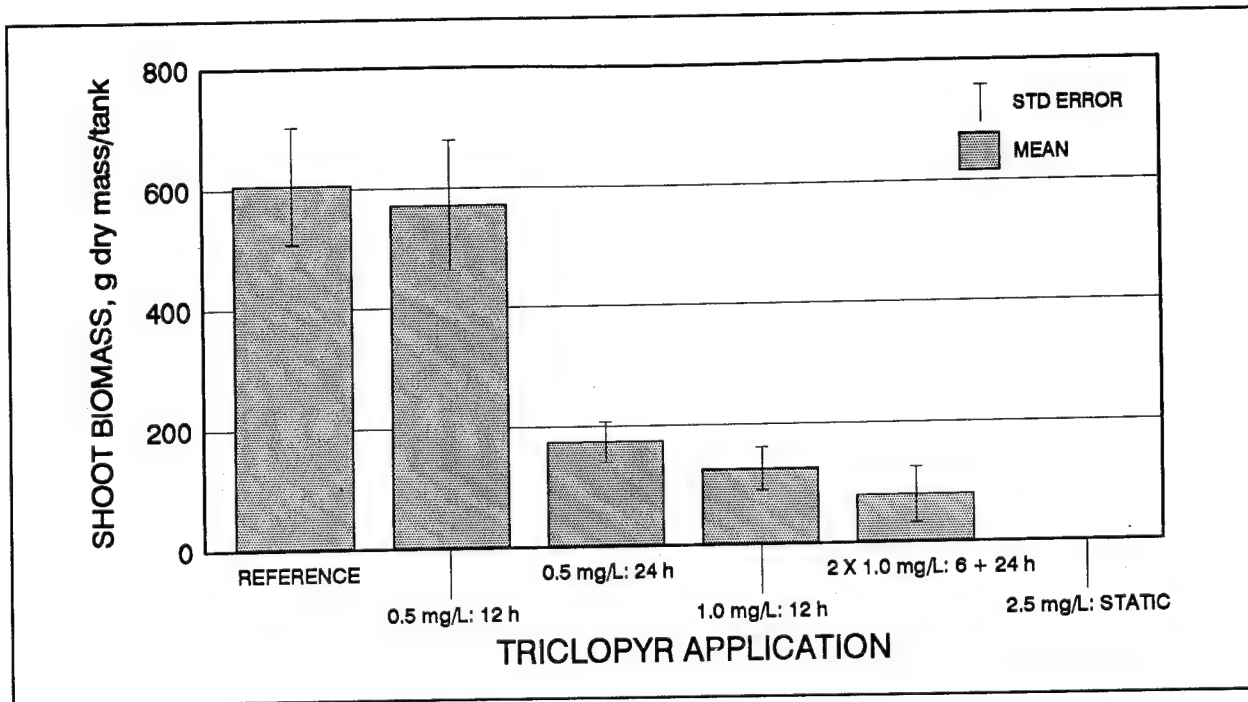


Figure 2. Shoot biomass (g dry mass/tank) of Eurasian watermilfoil measured at the end of the experiment. Values are means and standard errors based on three replicate tanks per treatment, except for the static 2.5 mg/L treatment which was unreplicated

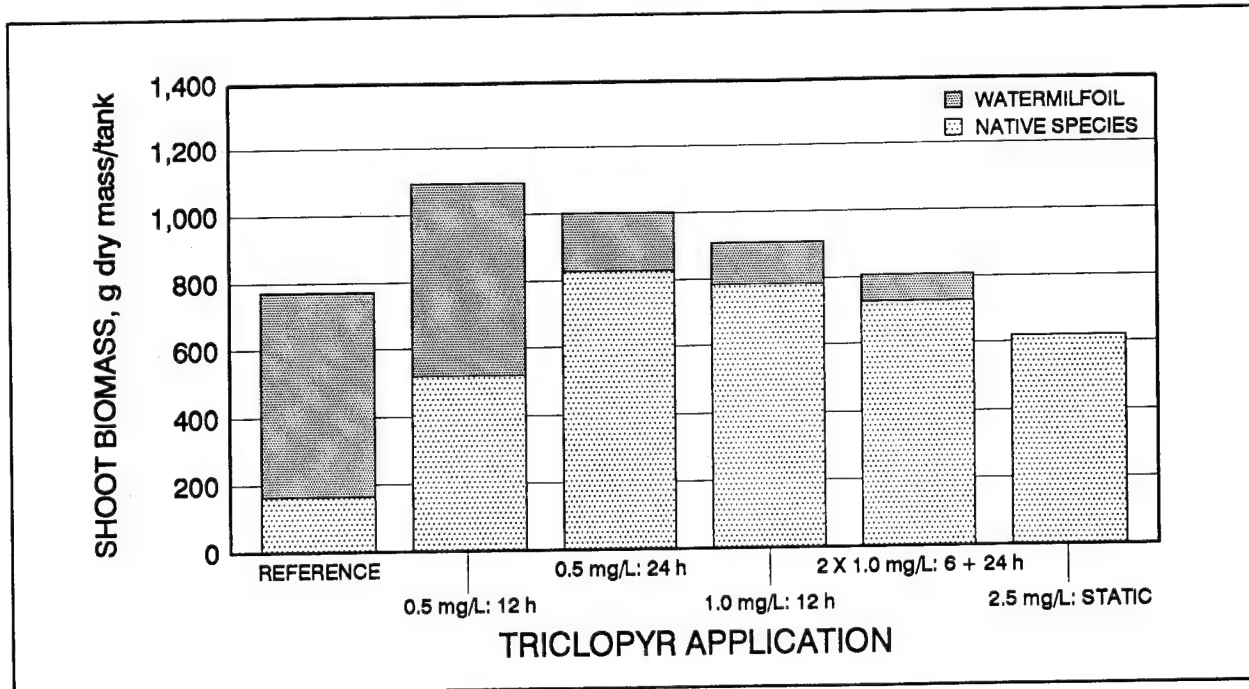


Figure 3. Total community shoot biomass (g dry mass/tank) measured at the end of the experiment. Community biomass includes that of Eurasian watermilfoil as well as the sum of all native species in the experimental communities. Values are means and standard errors based on three replicate tanks per treatment, except for the static 2.5 mg/L treatment which was unreplicated

effective treatment. This reduction suggests that one or more of the native species may be susceptible to triclopyr when it is applied at full label rate.

Conclusions

Low concentrations (0.5 to 1.0 mg/L) of triclopyr are effective at controlling Eurasian watermilfoil even when exposure periods are short (24 and 12 hr, respectively). These treatments elicited only minimal adverse effects on native members of the community, and once the dominance of Eurasian watermilfoil was eliminated, native species in these communities flourished.

These results indicate that triclopyr can be used to restore plant communities impacted by an infestation of the exotic, Eurasian watermilfoil. A similar result has been observed following triclopyr treatment in the Pend Oreille River, Washington (Madsen, Getsinger, and Turner 1994).

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Field Evaluation of Triclopyr in Lake Minnetonka: An Overview (32404)

by
Kurt D. Getsinger¹

Introduction

The herbicide triclopyr (3,5,6-trichloro-2-pyridinyloxyacetic acid), manufactured by DowElanco, is being evaluated for the selective control of exotic weeds (primarily Eurasian watermilfoil, purple loosestrife, and waterhyacinth) in aquatic sites around the U.S. Field studies have been conducted using the triethylamine salt formulation of triclopyr (Garlon[®] 3A) under EPA Experimental Use Permit (EUP) labels in numerous states, including Alabama, Georgia, Florida, Michigan, Minnesota, and Washington since 1986. This herbicide has been labeled for forestry and other noncrop terrestrial use since 1978, and has recently been used under EPA specific exemption labels for purple loosestrife control in Minnesota and broadleaf weed control in rice in Arkansas, Louisiana, and Texas.

Researchers at the WES have evaluated triclopyr as an aquatic herbicide under laboratory and field conditions since 1982 (Getsinger and Westerdahl 1984; Green et al. 1989; Getsinger, Turner, and Madsen 1992; Netherland and Getsinger 1992). Results from these studies have shown that triclopyr can provide selective control of Eurasian watermilfoil with no adverse impact on nontarget organisms or water quality.

Triclopyr is a systemic herbicide which is readily absorbed by shoots of actively growing susceptible plants, disrupting cell elongation in a manner similar to other synthetic plant auxins (WSSA 1989). In submersed plant communities dominated by Eurasian watermilfoil, triclopyr can provide excellent control of milfoil while minimizing the impact on na-

tive submersed species such as elodea, pondweed, and wild celery. This selective control will encourage the re-establishment and growth of a diverse native plant community (Getsinger, Turner, and Madsen 1993, 1994; Madsen, Getsinger, and Turner 1994).

The primary degradative mechanism for triclopyr in aquatic environments is photodegradation, with oxamic acid as the principal by-product (McCall and Gavit 1986; Solomon et al. 1988; Woodburn et al. 1993). Triclopyr will also undergo biodegradation in aquatic environments, with 3,5, 6-trichloro-2-pyridinol (TCP) as the major degradation product (Lickly and Murphy 1987; Norris, Montgomery, and Warren 1987; Woodburn and Cranor 1987; Grant 1992). Triclopyr and TCP have little to no effect on sediment or water microorganisms, and show low toxicities to aquatic insects, shellfish, fish, and waterfowl (Hedlund 1972; Dow Chemical 1988; WSSA 1989). Results from large-scale field studies have shown that dissipation/degradation in water is rapid, with half-lives of less than 4 days, and that residue accumulation of triclopyr and TCP in sediment, plants, shellfish, and fish is negligible (Getsinger and Westerdahl 1984; Dow Chemical 1988; Green et al. 1989; Woodburn et al. 1993).

In an effort to verify previous laboratory and field dissipation characteristics of triclopyr in aquatic sites, the WES Chemical Control Technology Team directed and conducted a large-scale dissipation study of triclopyr in Lake Minnetonka, MN. Primary study cooperators included the Minnesota Department of Natural Resources, the CE St. Paul and Jacksonville Districts, the Lake Minnetonka Conservation

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District, the University of Florida Center for Aquatic Plants, DowElanco, and Braun Intertec.

The objectives of this study were to:

(1) establish dissipation curves for triclopyr applied at the proposed maximum label rate as the triethylamine salt to an aquatic environment, (2) follow the formation and decline of triclopyr's major metabolite, TCP, (3) establish residue levels of triclopyr and TCP found in nontarget organisms (fish, clams, crayfish, and plants), (4) assess the efficacy of triclopyr on Eurasian watermilfoil, and (5) document the recovery of the aquatic plant community following triclopyr application. Study objectives 1 through 3 were accomplished under EPA Guidelines 164-2 and 164-5 following Good Laboratory Practices requirements.

This article will present an overview of the Lake Minnetonka study protocol. Other articles in this proceedings (Fox and Haller 1995; Madsen and Getsinger 1995) will report on specific aspects of the study.

Study Site and Treatment Plots

Study site

The study site was located in eastern Minnesota at Lake Minnetonka near Minneapolis (Figure 1). Lake Minnetonka consists of 15 interconnected basins with a surface area of some 5,800 ha (14,000 acres). At normal full-pool elevation, the lake has a mean depth of 7 m and a maximum depth of 31 m. Grays Bay Dam, located on the northeastern shore of the lake, regulates water during periods of floods and low flows. Five major streams enter Lake Minnetonka from the west and north, with the major outlet being Minnehaha Creek below Grays Bay Dam.

The exotic submersed macrophyte, Eurasian watermilfoil, is widespread in the lake and occupies a total area of over 500 ha. Despite ongoing mechanical and chemical control programs, this aggressive weed is displacing the desirable native plant community. An ecological description of Lake Minnetonka and its watershed can be found in Smith, Barko, and McFarland (1991).

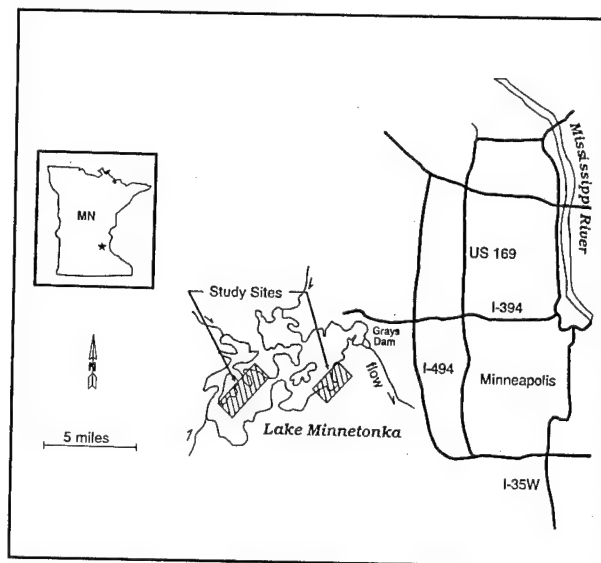


Figure 1. Triclopyr dissipation study site, Lake Minnetonka, MN

Treatment plots

Three 6.5-ha (16-acre) treatment plots were established in separate arms of the lake (Figure 2). Two of the plots were designated for triclopyr treatment and the third was used as an untreated reference (or control) plot. The vegetative communities located in these plots represented the types normally treated with herbicides in Lake Minnetonka. The stands were dominated by dense growths of the target weed, Eurasian watermilfoil, but also contained a sparse understory of nontarget native plants. Although not intended as replicates, these plots were selected for similarities in water depth and plant communities. The plots were separated by distances (5 to 10 km) great enough to preclude herbicide cross-contamination.

Plot A (mean depth = 2.1 m) was located along the west-northwest shore of Phelps Bay and was selected to receive a triclopyr treatment of 2.5 $\mu\text{g/L}$ (ppm) using a subsurface injection application technique. Plot B (mean depth = 1.9 m) was located in the southernmost protected arm of Carsons Bay and was selected to receive a triclopyr treatment of 2.5 $\mu\text{g/L}$ using a surface broadcast application technique. Plot C (mean depth = 2.5 m) was located along the northwest shore of Carman Bay and was chosen as the untreated reference

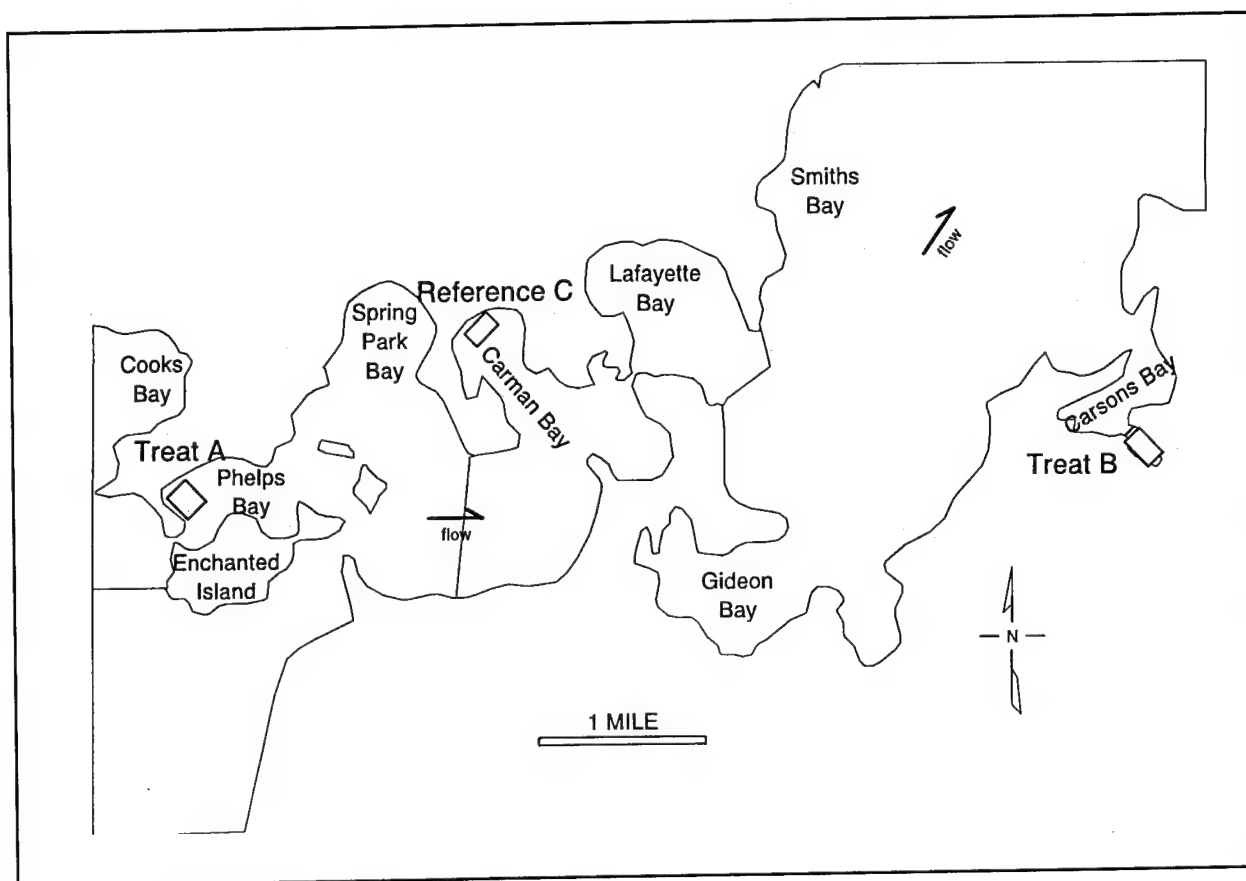


Figure 2. Treatment plots A and B and reference plot C at Lake Minnetonka study site

plot. Details of the triclopyr application procedure and pre- and 6-week posttreatment efficacy assessment are presented in Fox and Haller (1995) and Madsen and Getsinger (1995), respectively.

Plot design

A schematic of the basic plot design is shown in Figure 3. Each triclopyr-treated plot contained five internal sampling stations, one in the center of each quadrant and one in the center of the plot. External sampling stations were located 100 m from the edge of the plots (three at Plot A and one at Plot B), and at intervals of 400, 800, and 1600 m from the plot boundaries. The 400- to 1600-m stations were used to establish potential set-back distances with respect to triclopyr applications and potable water intakes. Additional external sampling stations were established to follow any off-target movement of triclopyr not covered by the predetermined external stations (Fox and Haller 1995). The internal and ex-

ternal sampling stations served as fixed locations for the collection of water, sediment, and plants. The internal center plot station served as a fixed location for sampling of

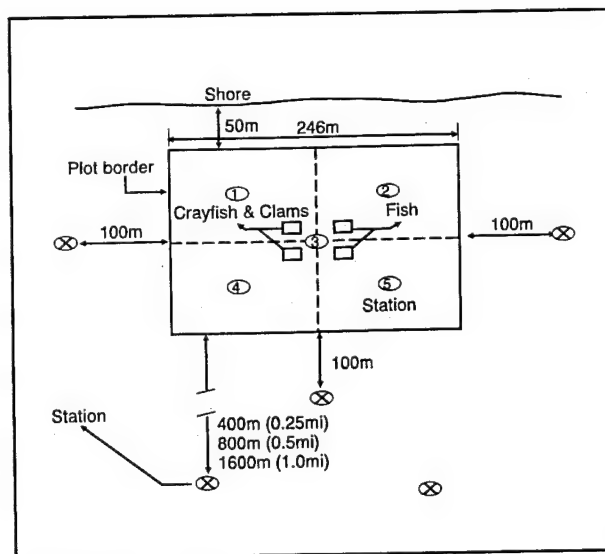


Figure 3. Schematic of triclopyr treatment plot and sampling stations

caged fish, crayfish, and clams. The corners of each plot, as well as each fixed sampling station, were signified for the duration of the study with standard mooring and can buoys, anchored to the bottom.

Data and Residue Collection

Meteorological conditions (air/water temperature, rainfall, humidity, wind, and light intensity) were measured using automated weather stations deployed on the shore adjacent to each plot. Water quality parameters (temperature, pH, dissolved oxygen, and conductivity) were collected using Hydrolab data sondes positioned at two water depths in the center of each plot. Water column profiles of light quality and quantity were measured using a LiCor submersible quantum sensor at selected locations within each plot.

Water exchange (flow), which could effect the off-target movement of triclopyr, was esti-

mated through the use of the fluorescent dye rhodamine WT (RWT). The RWT was tank-mixed and applied concurrently with the triclopyr in plots A and B. This tracer dye procedure also aided in establishing additional external water sampling stations following the triclopyr application, as mentioned above.

Triclopyr and TCP residue samples were collected for water, sediment, fish, crayfish, clams, and plants as shown in Table 1. Fish species included largemouth bass, bluegill, brown bullheads, and suckers. Residue samples were placed on ice immediately after collection, transported to frozen storage facilities, and air-freighted to the designated analytical laboratory.

Plant biomass and species diversity data were collected using a stratified-random sampling technique (Madsen 1993) by a CE-certified SCUBA dive team. Data were collected one week prior to triclopyr application and at 6 weeks posttreatment (Madsen and Getsinger

Table 1
Sampling Schedule for Triclopyr and TCP Residues

Schedule	Water	Sediment	Plants	Fish	Crayfish	Clams
Pre	x	x	x	x	x	x
1 hr	x		x	x	x	x
3 hr	x		x	x	x	x
6 hr	x	x	x	x	x	x
12 hr	x	x	x	x	x	x
1 day	x	x	x	x	x	x
2 day	x					
3 day	x	x	x	x	x	x
5 day	x					
7 day	x	x	x	x	x	x
14 day	x	x	x	x	x	x
21 day	x	x	x	x	x	x
28 day	x	x	x	x	x	x
42 day	x	x				
180 days		x				

1995). Additional data will be collected at 1-year posttreatment.

Preliminary Results

Preliminary results indicate that this triclopyr study will be a complete success with respect to meeting its original objectives. Critical information will be obtained on the dissipation of triclopyr and TCP in various compartments of the aquatic environment, and correlations between the behavior of RWT and triclopyr in natural waters will be verified and strengthened. Furthermore, triclopyr has continued to prove itself as an effective agent for the selective control of Eurasian watermilfoil.

Finally, results from this Lake Minnetonka study will be used to supplement the aquatic registration data package on triclopyr that will be submitted to EPA for technical review in the near future.

Acknowledgments

Many individuals from numerous organizations were involved in contributing to the success of this study. Credit must go to all of them, with special gratitude to D. Petty, C. Welling, G. Strommen, B. Haller, G. Gallagher, K. Wilkinson, K. Langeland, D. Keyes, J. Madsen, J. Skogerboe, G. Dick, A. Fox, J. Troth, M. Netherland, J. Reeves, M. Bull, C. Mayfield, K. Kretsch, T. McNabb, S. Farone, the Moonan Bros., M. Smart, M. Stewart, J. Everett, L. Nelson, P. Havens, M. Glenn, N. Flint, P. River, G. Rowley, H. Krosch, M. Jones, D. Baker, T. Mester, J. Olson, T. Kirby, H. Johnson, E. McNally, D. O'Borsky, N. Paurus, V. Ulrich, S. Rosenbloom, Y. Yue, E. Lewin, M. Melichar, G. Turner, and J. Brazil.

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Triclopyr/Dye Dissipation in Water: Lake Minnetonka

by

Alison M. Fox¹ and William T. Haller¹

Introduction

As a part of the registration requirements for obtaining an aquatic-use label for the herbicide triclopyr, a large-scale residue dissipation study was conducted in Lake Minnetonka in June 1994, as described by Getsinger (1995). In such studies, samples are collected both within and outside the treated plot so that the distribution of residues in water and, principally, their rate of dissipation may be estimated. Such data on the persistence of residues in the water column are used by the EPA to predict expected uptake by other organisms, and will influence the duration of water-use restrictions that may be imposed on the label. The herbicide manufacturer is also interested in the maximum distance at which detectable residues are found from the treated plot. Such information can be used to determine the minimum distance that applications can be made from a potable water intake, another important specification for the final product label.

In all previous herbicide registration studies the sites and times for sample collection have been predetermined, and usually any effects of lateral water movement (that were not detectable by conventional flow meters) have not been apparent until the expensive procedures of collecting and analyzing all samples have been completed. As previously noted (Fox and Haller 1994), water movement in herbicide treatment plots has been efficiently monitored in the field by tracking the distribution of the concurrently applied, conservative fluorescent dye, rhodamine WT. In the residue study for bensulfuron methyl registration conducted in Lake Seminole in 1989, rhodamine WT was applied concurrently with the herbi-

cide to two of the three treated plots (Fox, Haller, and Getsinger 1992). Use of the dye was important in that study in explaining the rapid rates of herbicide dissipation from the three plots (dye half-lives ranged from <1 to 4.4 hr).

The use of rhodamine WT in the Lake Minnetonka study differed from that in Lake Seminole in three important respects:

- (1) Dye dissipation tests conducted in Lake Minnetonka in August 1993 were used to select application sites in which the least water movement was likely.
- (2) The application and detection of dye were conducted under the same procedures of Good Laboratory Practice (GLP) as used for the herbicide residues. Thus, the dye data can be submitted to EPA as part of the registration package, and could be used, if necessary, to explain the effects of water movement on herbicide residue dissipation.
- (3) Not all sampling stations outside the plot were predetermined. By writing the study protocol to allow the addition of sampling stations after the direction of dye movement had been established, it was possible to optimize the collection of residue samples in relation to water movement from the plot.

The triclopyr residue data are not expected until mid-1995 so this paper will be limited to a discussion of the dye data collected in the field. Specific objectives of this paper are to describe the stability of the plots and to predict the likely vertical and lateral distribution of herbicide residues.

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Materials and Methods

The two treated plots (Phelps and Carsons Bays) have been described by Getsinger (1995). Phelps Bay was treated on June 21, 1994, with triclopyr and rhodamine WT at rates calculated to result in average concentrations of 2.5 mg/L and 12 µg/L, respectively. These materials were tank-mixed and applied from weighted hoses of three different lengths trailing from airboats. It took two airboats approximately 2 hr to treat the 6.5-ha (16-acre) square plot in Phelps Bay.

Water samples and dye measurements were collected from the five predetermined sampling stations within the plot at the following time intervals after the treatment of the respective section of the plot: 1, 3, 6, 12 hr and 1, 2, 3, 5, 7, 14, 21, 28, and 42 days. Sampling stations outside the plot (6 to 15, Figure 1a) were sampled, after the completion of the whole application, at the same time intervals, except that the 1-hr samples were not collected from the stations 200, 400, 800, and 1,600 m away from the plot.

Duplicate water samples of 400 ml were collected from the fluorometer outflow hose at the same time as the dye concentration and water temperature were recorded. Dye concentrations were subsequently corrected for temperature variations. Water was pumped through the fluorometer using 12-volt submersible bilge pumps (37.8 L/min = 10 gal/min). Within the plot these pumps were lowered to depths of 25 cm above the substrate, middepth, and 25 cm below the water surface. Outside the plot, samples were collected from the same three-depth profile as was found at the deepest of the five within-plot stations. Drinking-water quality PVC hoses were used and hoses and pumps were replaced daily, in addition to being exchanged between stations where dye was, or was not, detected.

The whole of Carsons Bay (approximately 6.9 ha = 17 acres) was treated on June 23, 1994, at rates of 2.5 mg/L and 10 µg/L of triclopyr and dye, respectively. These were applied using Radiarc sprayers mounted on the airboat bows. Water sampling procedures were identical to those in Phelps Bay, except

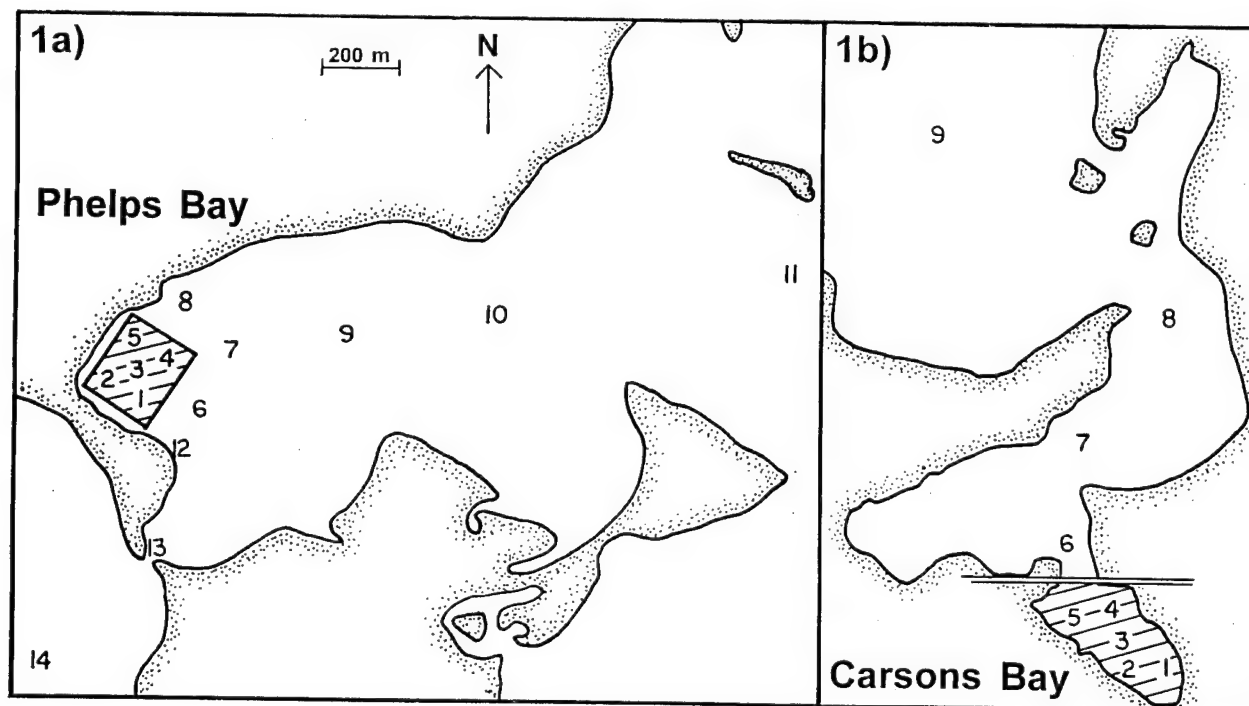


Figure 1. Maps of Phelps (1a) and Carsons (1b) Bays showing shorelines (stippled), treated plots (right hatch), and numbered dye/water sampling stations. Sampling station 15 in Phelps Bay is a straight line with stations 13 and 14, 800 m beyond station 14

that there was only one sampling station 100 m from the plot (Figure 1b).

Results and Discussion

Plot stability

One major advantage of adding dye to the herbicide was that it was possible to immediately see exactly where these materials were applied and how they moved. Although visible from the lake level, aerial photographs were spectacular in showing the precise alignment of the swaths and their diffusion together over the first few hours after application. Such photographs of the dye in Phelps Bay also clearly showed the outline of the plot which generally remained well-defined and stable for several days.

This visual portrayal of plot stability in Phelps Bay was reflected in the relatively long dye half-life of 94 hr, which had an r^2 value of 0.93 indicating a good fit of the data (averaged from the five within-plot stations) to the exponential dilution model. The dye half-life in Carsons Bay was 152 hr ($r^2 = 0.98$), a larger value that reflects the more isolated and dead-end nature of this plot. These are long dye half-lives compared to those found in many lake studies (Fox and Haller 1994). They are also substantially longer than the 51.2-hr dye half-life found after concurrent treatment of rhodamine WT with triclopyr in a cove on the Pend Oreille River (Turner, Getsinger, and Netherland 1994).

In that study, with sampling up to 7 days after treatment, dye and triclopyr half-lives were not significantly different. It is predicted that over the 6 weeks duration of the Lake Minnetonka residue sampling, triclopyr half-lives will be shorter than for the dye, due principally to photo/microbial degradation of the herbicide and its uptake by the vegetation. Dye was not detected in any samples collected 6 weeks after treatment and it is likely that triclopyr residues disappeared prior to that.

The most important issue relating to the dye half-lives is that even if the herbicide shows a very short persistence, the dye data

indicate that this cannot be attributed to unpredictable water movement. This provides a significant validation of the whole residue study. Had the dye half-lives been very short (e.g., <8 hr) the pertinence of such a study could have been questioned and it might have been necessary to abort the program of sampling and analysis so that the study might be repeated at a more suitable site. In such a case, the early abandonment of an inappropriate study could save considerable amounts of time and money, a saving that would not be possible if the dye were not used.

The y-intercepts of the dye dilution curves, 11.2 $\mu\text{g/L}$ in Phelps and 8.8 $\mu\text{g/L}$ in Carsons, indicated that the objective concentrations of 12 and 10 $\mu\text{g/L}$, respectively, had been closely achieved.

Vertical dye distribution

A comparison of the top and bottom water samples within the plot showed differences in the vertical distribution of dye between the two plots (Figure 2). With the exception of the first sampling time, the dye concentrations at the surface and bottom of the water column in Phelps Bay were generally similar, becoming almost identical by 3 days after treatment. The disparity between dye concentrations at these depths in Carsons Bay was much greater, especially within the first 24 hr, and a uniform vertical distribution was not obtained until 5 days after treatment.

These differences between the plots can be attributed to the respective application methods. The weighted hoses were used in Phelps Bay with the intention of penetrating any thermocline, and of distributing the dye and herbicide into the Eurasian watermilfoil stands. The use of Radiarc sprayers in Carsons Bay was intended to simulate the type of surface treatment that might be expected from a hand gun or an aerial application. The greater concentrations of dye at the water surface indicate that this simulation was effective. Even though this application was started early in the morning, surface water temperatures (average 25.1 $^{\circ}\text{C}$) were 0.5 $^{\circ}\text{C}$ higher than those at the lake bottom, a difference which had increased

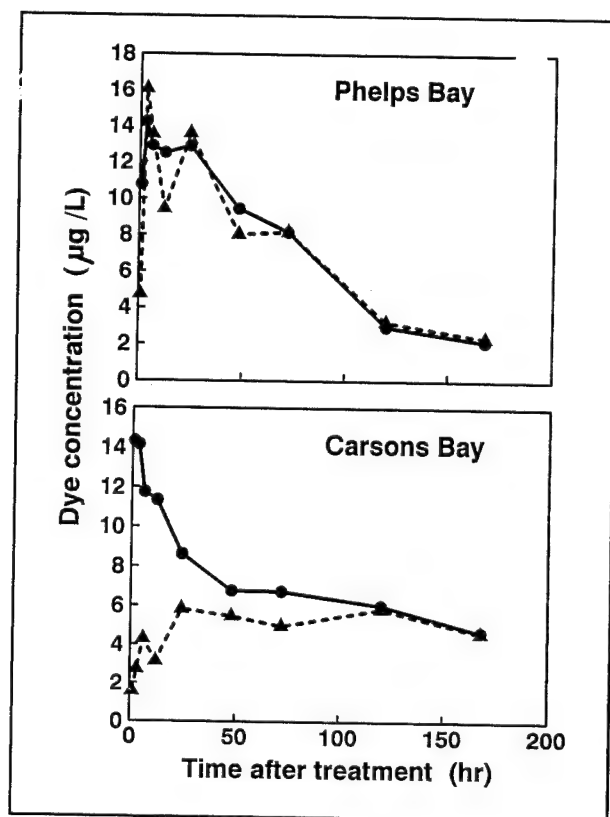


Figure 2. Dye concentrations up to 7 days after treatment at the top (solid line) and bottom (dashed line) of the water column averaged for all sampling stations within each plot

to 1.1 °C by the end of the first day. Although these temperature differences appear very small, under the relatively windless conditions that were present at the time of treatment, they may have been sufficient to maintain enough of a thermal stratification to reduce the rate of vertical mixing of the dye.

Lateral dye distribution within treatment plots

Within 2 days of the application to the Phelps Bay plot, it became apparent from aerial observations that the dye was disappearing from just the southeast corner of the plot (nearest sampling station 4, Figure 1a). This sudden reduction in dye concentration was evident from a comparison of average dye data from station 4 with that averaged for the other within-plot stations (Table 1). In Carsons Bay, dye concentrations at the two stations (4 and 5) nearest the mouth of the bay were generally lower than at the other stations (Table 1).

It is possible to surmise many reasons for these variations in lateral distribution of dye within the plots (e.g., differences in water depths, plant density, peripheral water

Table 1
Dye Concentrations (µg/L) Averaged Over All Depths For Sampling Stations Within the Treated Plots Showing Relative Reductions at Station 4 in Phelps Bay and Stations 4 and 5 in Carsons Bay

Time After Application	Phelps Bay Stations 1,2,3,5	Phelps Bay Station 4	Carsons Bay Stations 1,2,3	Carsons Bay Stations 4,5
1 hr	8.7	12.2	8.8	9.5
3 hr	16.4	12.8	10.8	7.9
6 hr	12.7	14.6	11.0	5.6
12 hr	11.5	10.6	9.3	6.0
1 day	13.8	10.4	8.9	5.5
2 day	10.3	3.3	7.2	5.1
3 day	9.5	2.4	7.0	4.7
5 day	3.8	0.5	6.9	4.6

movement). However, the important issue is that any similar lateral or vertical variations found in the distribution of the herbicide residues can, because of these dye data, be related to physical factors of uneven mixing or water movement. If only herbicide data were available, not only would the mixing and movement of water be speculative, but variations in factors that influence herbicide degradation (e.g., light penetration, proximity to microbial sources) would need to be considered.

Lateral dye distribution outside treatment plots

Dye was not detected at stations outside either plot within the first 3 hr after treatment. Although low concentrations of rhodamine

WT ($<0.5 \mu\text{g/L}$) were detected 6 hr after treatment at station 6 in Phelps Bay (Table 2), most of the water movement out of this plot was observed to be along the southwest shoreline, away from any of the predetermined sampling stations. The protocol flexibility and the in situ detection of the dye made it possible to add another transect of four sampling stations (12 to 15, Figure 1a) in the direction in which the dye was seen to move. The $2.8 \mu\text{g/L}$ dye concentration at the water surface of station 12 (100 m from the plot) 6 hr after treatment was greater than any concentration detected at the predetermined sampling stations outside the Phelps Bay plot (Table 2).

Dye had been detected at all sampling stations within 100 m of the Phelps Bay plot

Table 2
Time of First Dye Detection at Sampling Stations Outside the Plots with Maximum Dye and Predicted Triclopyr Concentrations

Distance From Plot (m)	Sampling Station	Days After Application Dye First Detected	Maximum Dye Detected: Concentration and Time		Maximum Potential Herbicide (mg/L)
			(µg/L)	(days)	
Phelps Bay					
100	6	0.25	1.72	0.5	0.38
	7	1	0.30	14	0.07
	8	2	0.82	7	0.18
	12	0.25	4.59	5	1.02
400	9	7 ¹	0.24	7	0.05
	13	0.5	0.41	2	0.09
800	10	7	0.15	14	0.03
	14	14	0.10	28	0.02
1,600	11	14	0.13	14	0.03
	15	14	0.07	21	0.02
Carsons Bay					
100	6	0.5	1.71	0.5	0.48
400	7	0.5	0.33	14	0.09
800	8	14	0.12	14	0.03
1,600	9	14	0.07	14	0.02

¹ 0.01 at surface only, 1 day after treatment (dat.); 0.03 at surface only, 3 dat.

by 2 days after treatment but concentrations at stations 7 and 8 never exceeded 1 µg/L (Table 2). Dye was detected within 12 hr at station 13 (400 m from plot) but was not significantly detected at station 9 until 7 days after treatment (Table 2).

In Carsons Bay, it took 12 hr for dye to reach station 6, 100 m from the plot. Low dye concentrations (<0.1 µg/L) were detected at station 7 at the same time but dye was not detected again at this distance from the plot (400 m) until 7 days after treatment (Table 2). Although dye concentrations were measured across the bay between stations 6 and 7, there did not appear to be water movement from the plot that was likely to be missed at the pre-determined sampling stations, so no further stations were added.

Dye appeared to be present at all sampling stations in both bays between 14 and 28 days after treatment, but concentrations above 0.15 µg/L were not detected at sampling stations beyond 400 m from the plots (Table 2). Dye was not detectable at any sampling stations by 42 days after treatment.

Because of the processes of triclopyr degradation outlined above, it is likely that herbicide residues will not be found in any of the later samples that contained dye (21 to 28 days). Also, the low dye concentrations detected at stations 800 and 1,600 m outside the plots may have resulted from changes in background fluorescence throughout the lake (e.g., algae bloom, increased suspended sediments). While such interference is usually minimal with rhodamine WT, its presence may be considered a possibility in situations in which no zero dye values are detected. Readings taken well away from any likely influence of the plot that showed zero at these times would tend to support the validity of data from the sampling stations, but such readings were not collected. Thus, it is quite possible that no herbicide will be detected at sampling stations over 400 m from the plot but this cannot be confirmed until the triclopyr residues are analyzed.

A more accurate prediction may be made in relation to maximum potential herbicide

concentrations outside the plot, assuming a sustained 1:1 ratio between dye and triclopyr concentrations. Based on the conservative estimates of initial dye concentrations in the plots derived from the y-intercepts of the dilutions curves described above, initial concentration ratios of herbicide:dye can be assumed of 2,500:11.2 in Phelps Bay and 2,500:8.8 in Carsons Bay. Using these ratios the maximum predicted herbicide concentrations outside the plots can be calculated from the maximum dye concentrations (Table 2).

The potable water tolerance for triclopyr is anticipated to be 0.5 mg/L. The only stations with dye concentrations that might be predicted to have herbicide concentrations approximating or exceeding that value would be stations 12 in Phelps Bay and 6 in Carsons Bay. These predictions conservatively assume that there was no herbicide degradation and that the initial herbicide concentration of 2.5 mg/L triclopyr was achieved, even though dye concentrations were slightly lower than anticipated. Thus, it may be predicted with some confidence that although triclopyr residues may be detected at some sampling stations over 100 m from the plot, none will exceed the anticipated potable water tolerance. Such results predict that in lakes, application set-back limits might need to be no further than 400 m (¼ mile) from a potable water intake.

Summary and Future Work

The use of rhodamine WT in these types of large-scale residue studies has been shown to provide several advantages.

- (1) The demonstration that the plots remained stable throughout the study provided evidence that there was a satisfactory exposure of sediments and organisms to the herbicide, justifying the continuation of residue collection and analysis.
- (2) By comparing these rates of dye dissipation in the plots with dissipation rates calculated for comparable triclopyr residues, it will be possible to estimate

the influence of degradation processes on the persistence of triclopyr in the water column.

- (3) In the case of a plot from which water could move in various directions (e.g., Phelps Bay) the ability to track the dye allowed the placement of some sampling stations outside the plot within the known direction of water movement. Thus, the maximum rate and amount of herbicide residue movement outside the plot can be confidently estimated for such a site.

From a pragmatic perspective, the immediate confirmation that the herbicide had been efficiently dispersed and remained in the plot, provided a significant boost in morale of test personnel and a renewed motivation to continue with such an extensive and expensive study. The herbicide residue results are awaited with eager anticipation and data analysis will include correlations with the dye and comparisons of predicted and actual triclopyr concentrations. Because of these satisfactory estimations of dye dissipation, further studies in the triclopyr registration process can be planned and initiated with greater confidence than if the plot stability, and hence study validity, were still to be revealed by the residue analyses.

Acknowledgments

Although, as indicated by Kurt Getsinger, many people were involved in this study, the

following persons are to be specifically thanked for their involvement in the application and sampling of the dye and herbicide residues in water: George Gallagher and Kevin Wilkinson (USACE Jacksonville); Mike Bull, Mike Netherland, and John Skogerboe (WES); Margaret Glenn, Ken Langeland, and Jan Miller (University of Florida).

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Assessment of Aquatic Plants Before and After a Triclopyr Treatment in Lake Minnetonka, Minnesota

by

John D. Madsen¹ and Kurt D. Getsinger²

Eurasian watermilfoil (*Myriophyllum spicatum*) is an aggressive nonnative aquatic plant that has extensive impact on freshwater resources throughout much of the United States. The dense surface canopy formed by this species in up to 5 m of water may impede commercial navigation and recreational uses, and may reduce flood storage capacity of reservoirs and hydraulic efficiency of channels. Fragments may clog water outlets and power generation turbines. In addition to these impacts to human activities, dense Eurasian watermilfoil stands may degrade water quality. If this species invades native aquatic plant communities, it has been documented to reduce native plant growth and diversity, establishing a near-monoculture (Madsen et al. 1991; Madsen, Hartleb, and Boylen 1991).

By selectively removing these dense stands of Eurasian watermilfoil using herbicides, a native plant community can be restored which increases biodiversity. In a previous study, we documented that a treatment with the herbicide triclopyr (as Garlon[®] 3A) in the Pend Oreille River, WA, effectively controlled Eurasian watermilfoil, thereby increasing native plant abundance and diversity for up to 2 years after treatment (Getsinger, Turner, and Madsen 1994; Madsen, Getsinger, and Turner 1994).

In the present study, the primary purpose of the treatment was to perform a dissipation study of triclopyr within two sites in Lake Minnetonka, MN. Therefore, this treatment represents a worst case scenario in which the full label rate of triclopyr (2.5 mg L^{-1}) is used under low water exchange conditions.

Materials and Methods

Study site

Lake Minnetonka, located just west of the metropolitan area of Minneapolis, MN, was first found to have Eurasian watermilfoil in 1986. Eurasian watermilfoil had become a considerable nuisance in some locations by 1989 (Smith, Barko, and McFarland 1991); however, it has not dominated all littoral zone areas of the lake. Two treatment sites (Phelps and Carsons Bays) and one reference site (Carman Bay) were examined. Additional information on these sites is given in other papers within this proceedings (Getsinger 1995; Fox and Haller 1995).

Methods

In the center of each plot, five 100-m transects with marked intervals every 1 m were deployed perpendicular to shore, spaced 25 m apart. SCUBA divers recorded the plant species present in each 1-m interval of the transect to determine species distribution and diversity. In addition, four biomass samples were collected in a stratified-random manner from each transect using a 0.1-m^2 quadrat. These samples were shipped to the LAERF laboratory, where shoots were sorted to species, dried at 55°C , and weighed. All three sites were evaluated 1 week before treatment (pretreatment) and 6 weeks posttreatment. Biomass and diversity (as average number of species per transect interval) were analyzed statistically using a student's T-test. Transect

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frequency (e.g., percent cover) was analyzed using a chi-square test on a two-by-two comparison table of pre- and posttreatment values of intervals with and without the plants in question.

Results and Discussion

Species present

A total of 21 species were observed from quantitative samples (Table 1). Of these, 13 were monocots, 6 were dicots, and two were macroalgae (charophytes). Also, all but two were native species. Using a sum of all tran-

sect data, two of five most common species were nonnative submersed plants (curlyleaf pondweed, 25 percent; Eurasian watermilfoil, 55 percent). The native dominant species were coontail (56 percent), elodea (18 percent), and flatstem pondweed (18 percent).

Biomass

Eurasian watermilfoil biomass was substantially higher at the reference site (300 g m^{-2}) than at the two treatment sites (50 g m^{-2}) before treatment (Figure 1). However, biomass at the reference site did not significantly change, while a significant reduction in biomass

Table 1
Plant Species Observed in Quantitative Samples from Lake Minnetonka

Species	Common Name	Native (N) or Exotic (E)	Dicot (D) or Monocot (M)	Transect Frequency of Occurrence
<i>Ceratophyllum demersum</i>	coontail	N	D	56.0
<i>Chara</i> sp.	muskgrass	N	*	1.0
<i>Elodea canadensis</i>	elodea	N	M	18.0
<i>Heteranthera dubia</i>	water stargrass	N	M	3.0
<i>Myriophyllum sibiricum</i>	northern watermilfoil	N	D	0.1
<i>M. spicatum</i>	Eurasian watermilfoil	E	D	55.0
<i>Najas minor</i>	bushy pondweed	N	M	2.0
<i>Nitella</i> sp.	nitella	N	*	5.0
<i>Nymphaea odorata</i>	white waterlily	N	D	0.5
<i>Potamogeton amplifolius</i>	wideleaf pondweed	N	M	3.0
<i>P. crispus</i>	curlyleaf pondweed	E	M	25.0
<i>P. obtusifolius</i>	narrow pondweed	N	M	5.0
<i>P. pectinatus</i>	sago pondweed	N	M	5.0
<i>P. praelongus</i>	muskyweed	N	M	0.1
<i>P. pusillus</i>	narrow pondweed	N	M	1.0
<i>P. richardsonii</i>	Richard's pondweed	N	M	2.0
<i>P. robbinsii</i>	Robbin's pondweed	N	M	1.0
<i>P. zosteriformis</i>	flatstem pondweed	N	M	18.0
<i>Ranunculus longirostris</i>	water crowfoot	N	D	1.0
<i>Utricularia vulgaris</i>	common bladderwort	N	D	3.0
<i>Vallisneria americana</i>	water celery	N	M	*

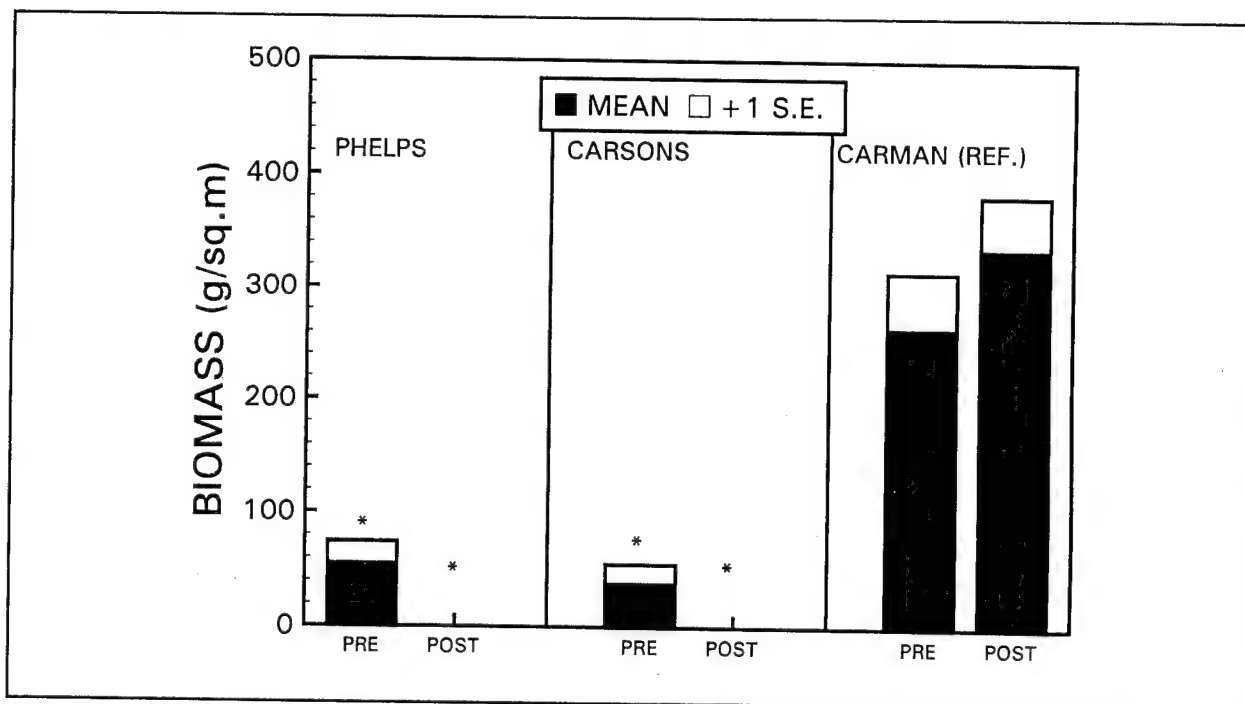


Figure 1. Biomass (g DW m⁻²) of Eurasian watermilfoil pre- (PRE) and posttreatment (POST) at the two treatment sites (PHELPS, CARSONS) and the untreated reference (CARMAN(REF)) site. Asterisks (*) indicate a significant difference between pre- and posttreatment at the $p = 0.05$ level using a T-test

occurred at both treatment sites—in fact, no Eurasian watermilfoil biomass was found posttreatment at either treatment site.

Native plant biomass did not change significantly at Phelps Bay after treatment (Figure 2). Phelps Bay had a higher exchange rate than Carsons Bay, resulting in lower concentrations of triclopyr over the period of the study. Carsons Bay retained higher concentrations of triclopyr, which may have resulted in some native plant mortality. Native plant biomass decreased significantly after treatment but was not completely eliminated. Carman Bay, with low native biomass before treatment, showed a significant increase in native plant biomass that may have been related to the senescence of the dense curlyleaf pondweed during this time period.

Transect data

Transect frequency data give better information on the distribution and diversity of plants than biomass data. Total plant cover did not change at the reference site but was

significantly decreased at both treatment sites (Figure 3). However, plant occurrence only decreased from 100 to 85 percent, which is far short of total plant eradication that is typically ascribed to aquatic herbicide treatments.

Eurasian watermilfoil distribution decreased significantly at both treatment sites, from approximately 70 percent before treatment to 0 percent after treatment (Figure 3). In fact, no rooted Eurasian watermilfoil was observed at either site after treatment, although floating Eurasian watermilfoil fragments were observed. In contrast, the distribution of Eurasian watermilfoil at the reference site actually increased.

Native plant coverage also decreased at both treatment sites, but only by 5 to 10 percent (Figure 3). In contrast, native plant coverage decreased from 60 percent in June to 30 percent in August at the reference site, without any herbicide application. The apparent contradiction with the increase in native plant biomass at this site may be explained through significant increase in biomass of a

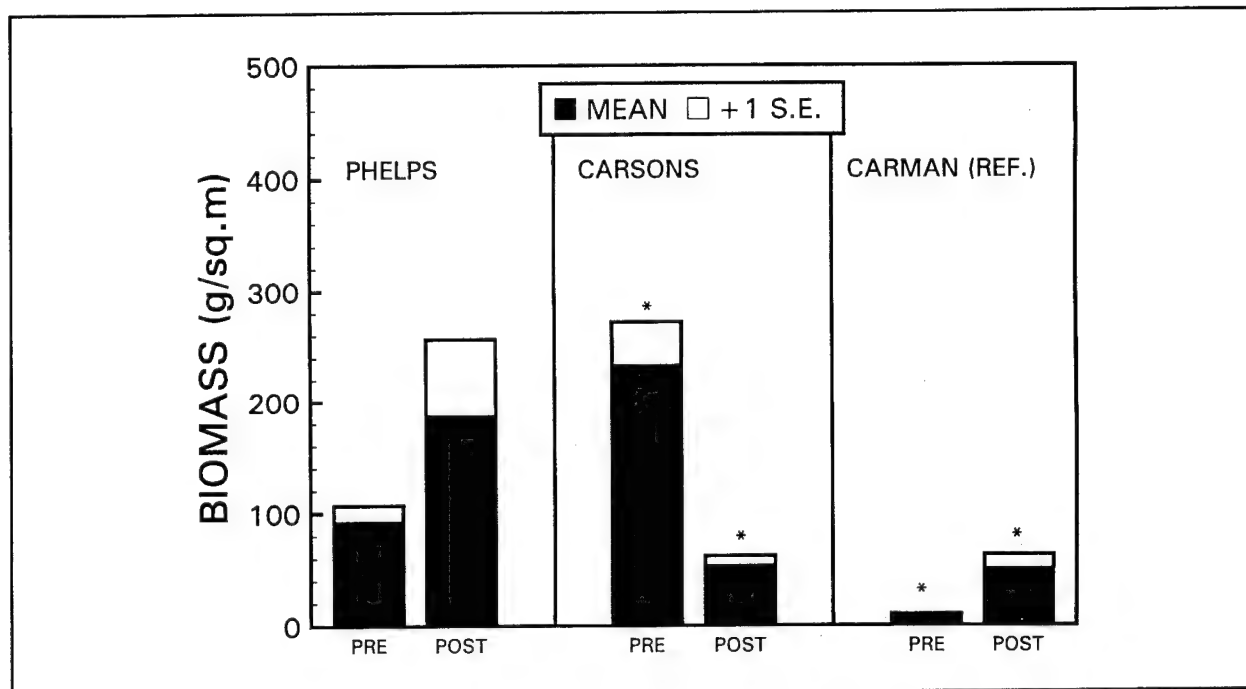


Figure 2. Biomass (g DW m^{-2}) of native plants pre- (PRE) and posttreatment (POST) at the two treatment sites (PHELPS, CARSONS) and the untreated reference (CARMAN(REF)) site. Asterisks (*) indicate a significant difference between pre- and posttreatment at the $p = 0.05$ level using a T-test

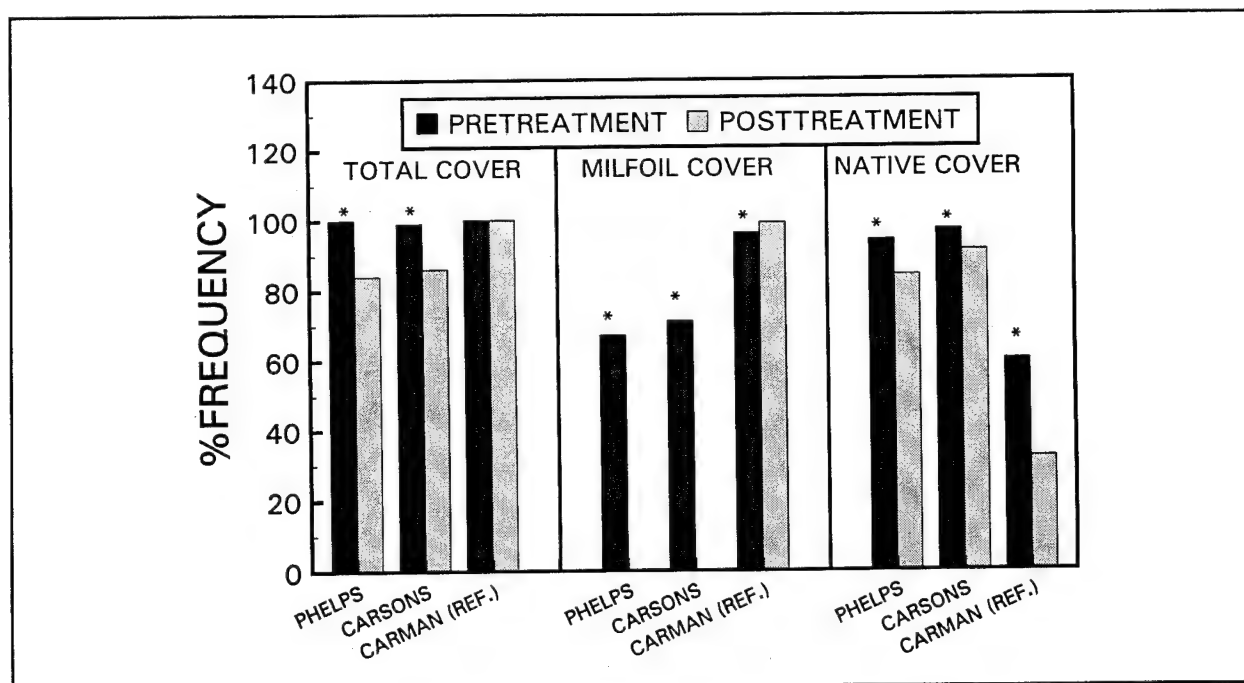


Figure 3. Percent frequency of observance along transects pre- (PRE) and posttreatment (POST) at the two treatment sites (PHELPS, CARSONS) and the untreated reference (CARMAN(REF)) site: total plant cover, Eurasian watermilfoil cover, and native plant cover. Asterisks (*) indicate a significant difference between pre- and posttreatment at the $p = 0.05$ level using a chi-square test on a two-by-two table

few species locally, and mortality of a few individuals that occurred throughout the bay. The decrease in native plant cover caused by the application of triclopyr was substantially less than observed in an untreated site.

Native plant diversity, as measured by average number of species per transect interval, decreased by almost 1.0 at both treatment sites (Figure 4). However, the diversity levels at the treatment sites were both above 1.0 after treatment, while the reference plot with dense Eurasian watermilfoil had average diversity of less than 0.5. Treatment with triclopyr at the full label rate may have caused the mortality of some native species. However, as was observed in the Pend Oreille, native plant diversity and abundance will probably increase substantially next growing season, after the removal of Eurasian watermilfoil biomass.

Conclusion

Treatment of triclopyr at the full label rate resulted in complete eradication of Eurasian

watermilfoil rooted plants in both treatment sites, as evidenced by biomass and transect data. Many native species survived, although some were affected by the treatment. Native species distribution and diversity both had small but statistically significant decreases. Native plant biomass was affected at one site, but not the other. However, it should be pointed out that native plant biomass, cover, and diversity remained higher after treatment than values for those parameters at the untreated reference plot, which had dense Eurasian watermilfoil.

Acknowledgments

R. Michael Smart, R. Michael Stewart, Joel Everett, and Michael Netherland were part of the CE SCUBA team collecting underwater data. Nicole Flint assisted in field data collection. Many others assisted both in the field and at the LAERF.

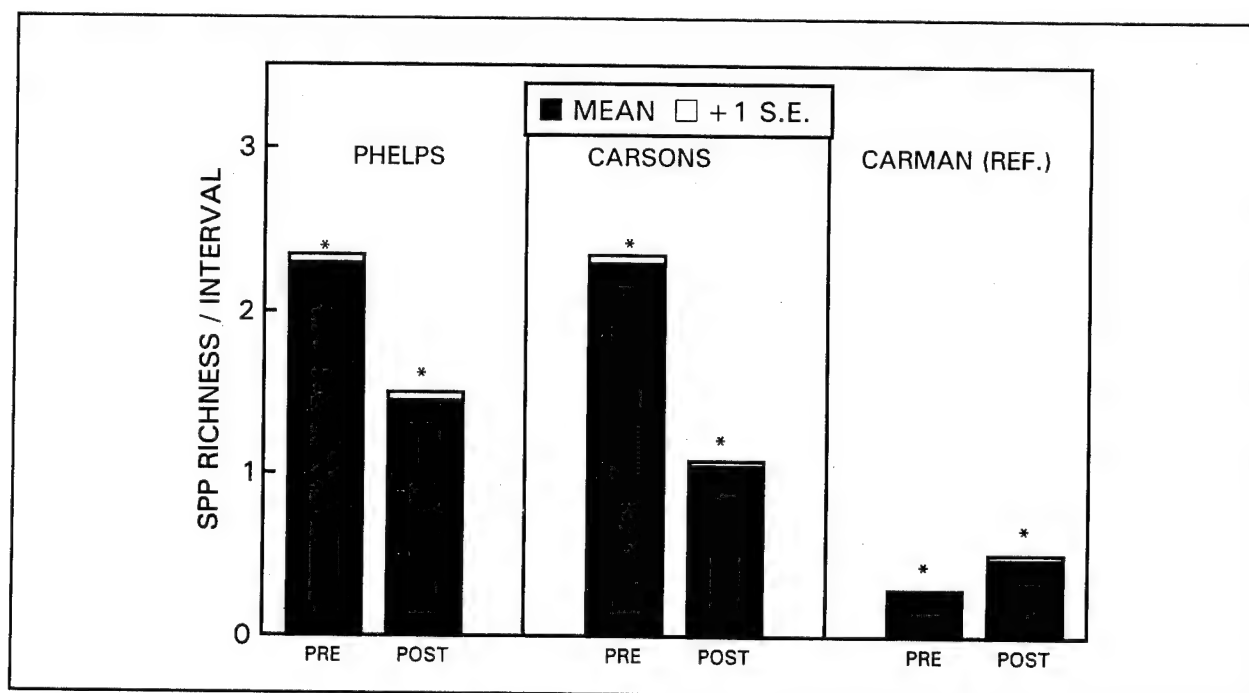


Figure 4. Species richness of native plants as number of species per transect interval pre- (PRE) and posttreatment (POST) at the two treatment sites (PHELPS, CARSONS) and the untreated reference (CARMAN(REF)) site. Asterisks (*) indicate a significant difference between pre- and posttreatment at the $p = 0.05$ level using a T-test

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Phenology of Aquatic Plants

by
John D. Madsen¹

The goal of phenological studies on aquatic plants is to determine points in the growing cycle of target nuisance species during which control tactics would be optimal, based on the storage of carbohydrates (Madsen 1993a,b). These "weak points" are then correlated to observable morphological indicators, such as flowering or fruit formation, thereby allowing the resource manager to easily identify the weak point. Weak points might include times of minimum stored carbohydrates during spring growth, called primary weak points, or times of reallocation of carbohydrates to overwintering storage structures, called secondary weak points.

During the past year, research concentrated on determining weak points for the nonnative species Eurasian watermilfoil (*Myriophyllum spicatum*). Research included sampling two pond populations to elucidate carbohydrate storage patterns in Texas populations and compare them with those found from literature sources in northern populations.

Methods

Research was performed at the LAERF in Lewisville, TX. During 1991, one pond population was sampled monthly, with 12 samples taken each month. During 1992 and 1993, six to twelve samples were taken from each of the two ponds, but the data will be combined in this presentation. Samples were sorted into root crowns, lower stems, upper stems, auto-fragments, and inflorescences and dried at 50 °C to constant weight. After grinding to pass a 1-mm grid, the samples were analyzed for total nonstructural carbohydrates at the CCTT laboratory at WES using previously de-

scribed analytical techniques (Madsen, Luu, and Getsinger 1993).

Carbohydrate Content of Eurasian Watermilfoil

Carbohydrates are primarily stored in the root crown and lower shoots of Eurasian watermilfoil. Primary weak points were observed in July of 1991, in April of 1992, and in May of 1993 (Figure 1). The earlier weak point would be more typical of a southern population, such as Texas. However, weak points can vary considerably from year to year and location to location (Table 1). Secondary weak points were observed in October of 1991, 1992, and 1993 for the Texas population (Table 1, Figure 1). Reallocation of carbohydrates to storage areas appears to increase as days shorten, even though water

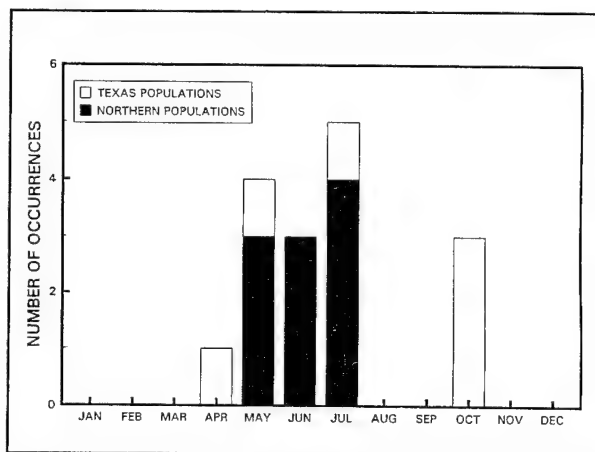


Figure 1. Frequency distribution of weak point timing by month of the year for Eurasian watermilfoil populations in northern areas and in Texas. Data from Table 1

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Table 1
Phenological Weak Points in the Seasonal Growth Cycle of Eurasian Watermilfoil
Populations as Determined by Minima in Annual Carbohydrate Storage

Lake	State/Province	Date of Weak Point	Citation
Buckhorn Lake	ON	06-15-79 06-15-80 06-15-81 05-30-82 07-15-83 07-15-84	Painter 1988 Painter 1988 Painter 1988 Painter 1988 Painter 1988 Painter 1988
Lewisville Ponds	TX	07-15-91 10-15-91 04-15-92 10-30-92 05-21-93 10-04-93	Madsen 1993a Madsen 1993a Madsen 1994 Madsen 1994 This Study This Study
Lake Washington	WA	07-30-80 05-30-81	Perkins and Sytsma 1987 Perkins and Sytsma 1987
Lake Wingra	WI	07-15-74 05-30-74	Titus and Adams 1979 Titus and Adams 1979

temperature is still within optimal growth conditions for Eurasian watermilfoil. By graphing the timing of weak points as presented in Table 1, it is apparent that northern populations exhibit only primary weak points, distributed evenly from May through July (Figure 1). The timing of primary weak points in Texas is split between April and July, but the secondary weak points are more uniformly distributed in October. This might suggest that fall treatments of Eurasian watermilfoil in southern populations, such as TVA reservoirs, could be employed to take advantage of a secondary weak point.

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Biological Control Technology

Review of Biological Control Research for 1994

by

Alfred F. Cofrancesco¹

Introduction

The Corps of Engineers (CE) has been involved in the management of aquatic plants since the late 1800s. The River and Harbors Act of 1899 directed the CE to remove aquatic vegetation that was hampering the operation of navigable waterways in Florida and Louisiana. Modifications to this act have, over the years, expanded the responsibility of the CE to include waterways influencing navigable waterways.

During the past 35 years the CE has conducted research on biological control of aquatic plants in conjunction with the USDA-ARS. Initially this research was focused on the control of floating plants. More recently we have become involved with submersed aquatic weeds, such as hydrilla and Eurasian watermilfoil.

This review will present information on research being conducted to identify insect and pathogen biological control agents. In FY94 there were six direct allotted work units and five reimbursable projects that addressed various aspects of biological control. This presentation will review the general approaches used in the development of biocontrol agents and will give an overview of the current research that is being conducted on hydrilla and Eurasian watermilfoil. Detailed information on particular research projects can be found in other reports of this proceedings.

Most problem plants in the United States, particularly in the aquatic and wetland environments, are exotic species. The plants (weeds) usually have been introduced into favorable environments without their natural enemies. These exotic plants have the ability to increase rapidly, outcompeting the native

vegetation for habitat and resources (Harley and Forno 1992). At present, the most successful biocontrol programs to manage aquatic plant growth in the United States have used insects from the native range of the problem plant. Generally, this is known as "Classical Biological Control."

The classical approach is based on the concept that the target plant has natural control agents present in its native range, and the introduction of these natural enemies will re-establish the pressure that the noxious plant normally experienced. In this approach, control agents (natural enemies) are introduced into areas that are not part of their native range to manage an introduced noxious plant (Harley and Forno 1992). In general these agents are host-specific arthropods, nematoda, or plant pathogens.

Biocontrol Agents

The process of introducing biocontrol agents can be divided into five phases—overseas surveys, overseas research, quarantine research, release and establishment, and technology transfer/resource management.

The first-phase surveys are conducted in the problem plant's native range, and potential biocontrol agents are identified. During the second phase, overseas research is conducted on potential agents to determine the agents' general specificity to plant species. In the third phase, insects are shipped into a United States quarantine facility where intensive host-specificity testing is conducted and studies are performed to determine other key preference factors of the agent. If the agent is approved for release, the USDA-APHIS-PPQ will issue a release permit. Once the release

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permit is received, the fourth phase is initiated and the agent is released into the field. Its population development and dispersal is monitored. The fifth phase consists of transferring the technology on how to employ and manage the biocontrol agent as a natural resource.

Although the five-step process appears simple, the tasks outlined can be difficult. In general, when a new biocontrol project begins for a noxious plant, it will take between 7 and 8 years before the first agent is released. This time frame may seem long; however, the introduction of an agent that may attack a non-target host could be devastating. Agricultural crops and the natural environment could be severely impacted, so great care is taken in the introduction process.

Biological control methods are worth the time and effort that are required for their development. They are extremely cost-effective. Once a host-specific agent is released and established, it generally will maintain itself in the environment and only minimal costs will be incurred to monitor and manage the population. As the population of the biocontrol agent develops, it will respond to the population growth of the noxious plant but will never completely eliminate the plant. Researchers attempt to reduce the population of the target plant below problem levels by introducing a complex of agents that attack various aspects of the plant or its life stages.

Research

Biocontrol research in FY94 has been conducted on submersed aquatic plants. Research has been conducted using both insects and pathogens to manage hydrilla and Eurasian watermilfoil. Hydrilla is a submersed aquatic plant that clogs waterways and impedes navigation (Schardt and Schmidt 1989). The plant was introduced into the United States by business as a plant for fish aquariums. The plant has spread rapidly throughout the southern United States and along the east coast as far north as Delaware. In addition populations of the plant are found in California. Research began in 1980 to determine the area of origin

for this problem plant. Surveys were conducted throughout Africa and Australia and parts of Asia (Balciunas 1982, 1983, 1984, 1985, 1987; Balciunas and Dray 1985). Biocontrol agents were most abundant in India and Australia. In 1984 the USDA established a research facility in Australia with the support of the CE.

Insects

The first insect released on hydrilla was a weevil (*Bagous affinis*) from Pakistan. Releases were made in 1987 in Florida (Center 1989; Center and Dray 1990); however this insect feeds on the tubers of the hydrilla plant when water has receded from the plants (Buckingham and Bennett 1994). While this situation is common in Pakistan it is very uncommon in Florida except when lakes are drained. This type of life cycle has made it difficult to establish field populations. This insect is being used in the canal systems in California which have annual periods of draw-down. Initially, problems occurred in establishing field protection; however, by mid-1990, field populations of the weevil were established in Florida (Center and Dray 1990).

The second insect released in the United States as a biocontrol of hydrilla is *Hydrellia pakistanae* an ephydrid fly from Pakistan. This insect was released in 1987 in Florida. The larvae mine the leaves and are the only life stage of the insect which impacts the plant (Buckingham and Bennett 1994). The life cycle is short—approximately 20 days (Center and Dray 1990). Insect populations have established and are widespread in Florida and are also present in Alabama, Louisiana, Texas, and California.

The third biocontrol insect released in the United States for hydrilla was *Hydrellia balciunas*, an ephydrid fly from Australia. This fly causes damage similar to that of *H. pakistanae*. The first field release of this insect occurred in September 1989 in Broward County, FL (Center, Cofrancesco, and Balciunas 1990). This insect has been established in Texas and is being distributed at other locations.

Additional insect biocontrol agents are being studied as biocontrol agents of hydrilla. A weevil from Australia (*Bagous hydrillae*) has been released in Florida, Georgia, and Texas and results are discussed in detail in other papers in this proceedings.

Pathogens

Pathogens have also been explored as biocontrol agents of hydrilla. A fungal pathogen associated with hydrilla populations in Texas has demonstrated potential as a biological control agent for hydrilla (Joye 1990). Studies on the impact of this agent and the development of a commercial formulation are discussed in Shearer (1995).

Myriophyllum spicatum is a submersed aquatic plant native to Eurasia (Godfrey and Wooten 1979). It is believed by some that the plant was introduced into North America in the early 1800s (Reed 1977); however, others believe it was not introduced until the 1940s (Couch and Nelson 1986). Since its introduction, it has been widely distributed throughout the United States. The plant causes major problems in the northern lakes, where extensive monocultures of the vegetation develop and restrict the use of the habitat and outcompete the growth of native vegetation. Eurasian watermilfoil is the most extensive aquatic problem plant in the United States. It has been reported from over 30 states. Biocontrol research on this plant dates back to 1967 with work in Yugoslavia. The major emphasis has been on pathogens to control this plant because many of the European insects were already found in the United States.

A fungus *Mycoleptodiscus terrestris* was isolated from plants in western Massachusetts prior to 1979 (Gunner 1983). Testing at the University of Massachusetts funded by the CE indicated that this pathogen had potential as a biocontrol agent of Eurasian watermilfoil.

A new research effort in pathogen biocontrol was begun in FY94 which will examine exotic pathogen biocontrol agents. Researchers from WES and the USDA conducted the first surveys for exotic pathogens of Eurasian

watermilfoil and hydrilla in Asia, and WES also contracted the International Institute on Biological Control to begin surveys in Europe for exotic pathogens of Eurasian watermilfoil.

Research was conducted to examine the decline in Eurasian watermilfoil in certain North American lakes where herbivorous insects have been found associated with declining watermilfoil populations (Painter and McCabe 1988; MacRae, Winchester, and Ring 1990; Creed and Sheldon 1991). The amount of impact which these herbivores may have contributed to the declines still has not been determined; however, there appears to be a strong correlation with the presence of insects and the loss of plant buoyancy. Adult and larval weevils have been observed feeding on Eurasian watermilfoil stems. The damage caused could have an impact on the ability of the plant to float or allow an entry point for natural saprophytic pathogens. Research is being conducted at Middlebury College, Middlebury, VT, and at the University of Minnesota.

Summary

Biocontrol research during 1994 has focused primarily on two target plants: hydrilla and Eurasian watermilfoil. The research was conducted on five continents and in cooperation with ten research laboratories. Conducting research projects on five continents is a difficult process and requires communication, cooperation, and coordination among all persons involved. The successes that have been achieved prior to and during 1994 point to the fact that the researchers involved in this technology area are extremely dedicated individuals. These researchers have coordinated with sponsors and operational personnel to ensure that the technology being developed will be useful and compatible with operational programs.

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The Application of Foundational Research in Biological Control Projects

by
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The analogy of a pipeline is often used to describe the processes involved in developing a biological control program, with each discrete step being represented by a segment of the pipe. This analogy is useful for visualizing how one phase of a project (i.e. segment of a pipe) leads into the next, as foreign exploration, for example, is a necessary prerequisite to quarantine studies. The pipeline concept is oversimplified, however, because it leads to the impression that these processes are linear and that the flow is unimpeded. Unfortunately, this is almost never the case. Every facet of a project is fraught with problems that require research to overcome. Oftentimes this research is not directly related to project objectives and might therefore seem somewhat esoteric. However, the project may not be able to move forward without it. We have dubbed these types of studies "foundational research," in that they provide a supporting knowledge base that is essential to the attainment of project objectives, even though the connection is not always obvious.

An excellent example is provided by ongoing Australian research. The Australian Commonwealth Scientific and Industrial Research Organization (CSIRO) is developing a biological control project against the invasive exotic plant *Mimosa pigra* (= *M. pellita*), a leguminous shrub or small tree of wetland habitats. *M. pigra* is native to the Neotropics and ranges from southern Texas to South America. *M. pigra* var. *pigra* was introduced into Australia in the late 1800s (Miller and Lonsdale 1987) where it persisted at innocuous levels for many years. Populations erupted in the late 1970s to dominate nearly 450 km² of seasonally inundated floodplains by the late 1980s

(Pitt and Miller 1988) and over 800 km² by 1994 (Forno, Heard, and Day 1994). The consequent changes in community structure have been disastrous to native organisms. Braithwaite, Lonsdale, and Estbergs (1989) regard this conversion of native sedgeland to tall shrubland as more disruptive, in terms of the effects on associated native fauna, than the replacement of native woodlands by introduced pine plantations.

The tremendous reproductive potential of this plant and the persistence of seeds in the soil account for its weedy nature (Lonsdale, Harley, and Gillett 1988). Seed banks in Australia may contain 12,000 seeds/m², two orders of magnitude higher than in its native range (Lonsdale, Harley, and Gillett 1988). It is obviously quite important to control seed production in order to manage this plant. CSIRO has introduced several *Mimosa*-feeding insects and is also developing plant pathogens as biological agents (Forno 1992). However, this example deals with two small weevils, *Coeloccephalapion aculeatum* (Fall) and *C. pigrae* Kissinger, both of which have the potential to destroy a high percentage of the flowers.

C. aculeatum severely damages developing inflorescences. Both the adults and larvae destroy flower buds. It was introduced with the expectation that, as a result, seed production would decrease (Forno, Heard, and Day 1994). However, flowers are not present during the long dry season prevalent in the Northern Territory, and it was predicted that *C. aculeatum*, being completely dependent on buds, would have difficulty persisting during this period. Fortunately, the Australians had appointed an ecologist to the biological control

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program who had studied the ecology of *M. pigra* in the afflicted region. They were therefore able to recognize these limitations on the effectiveness of *C. aculeatum* and sought additional agents with better capabilities for survival through the dry season. They shifted their exploratory studies to an area in Venezuela with a wet/dry season paralleling that of the Northern Territory and found a previous undescribed species of *Coelocephalapion*, later named *C. pigrae*. They have now released this species which produces damage to flower buds similar to that of *C. aculeatum* (Forno 1992). Unlike *C. aculeatum*, however, the adults also feed on leaves and can hopefully better survive the climate of the recipient region. This course was taken because of the knowledge project scientists had attained, both about the phenology of the plant in the adventive area and about the entomofauna associated with the plant in its native range. This type of feedback from a domestic program to an active foreign program can often be responsible for the ultimate success of a project. Unfortunately, as projects progress, funds must often be shifted from one phase to the next. It's therefore not always possible to maintain effort both at home and abroad simultaneously, but it is desirable to maintain some activity at the foreign end of the program.

Another example of the importance of foundational research is provided by a project on biological control of waterlettuce sponsored by the Jacksonville District Office of the Corps of Engineers (ACE-JAX). In anticipation of the release of biological control agents, we monitored several waterlettuce populations to establish baseline parameters for future efficacy studies, particularly with regard to phenology and population dynamics of the plant populations. Prior to initiation of this project waterlettuce was not known to produce seed in the United States. However, soon after beginning field studies, we discovered fruits on waterlettuce rosettes that contained mature seeds (Dray and Center 1989). Later germination trials revealed that most of these seeds were viable. Examination of one field site revealed the presence of a large seed bank containing nearly 5,000 seeds/m² (84 percent in the sediments, 16 percent in the mat). This,

compared to only 267 rosettes/m² on the water surface, indicates that typical control programs which target only the floating rosettes miss the 95 percent of the total population that is present as seed. Waterlettuce populations therefore recover rapidly after herbicide treatments. Although this discovery came about as the result of a biological control project, its implications are far broader. This capacity in the plant population to survive adverse conditions through the mechanism of seed reproduction certainly complicates all control strategies, including biological control. We previously thought that this plant would be easily controlled by one or two biological control agents, but we now believe that more will be required and that reproductive suppression must be an important objective.

Biosystematics also provides foundational research that is essential to biological control activities. Proper identification of the plant, possibly at the subspecies or cultivar level, is essential to ensure that candidate biological control agents have been collected from the correct host. A clear and accurate understanding of the systematic relationship between the target plant and other species provides critical information. This information is needed to determine which nontarget species might be at risk and should therefore be closely scrutinized in host-specificity studies. A similar understanding of the systematic relationships between a biological control candidate and related species and their collective hosts provides further insight into the risk of spillover onto nontargets, i.e. an allopatric plant species that hosts a close relative of the proposed biocontrol agent might become a second host for the candidate agent after the agent is released into its range. Hence, it's important to know about the host plants of all near relatives of any candidate biological control agent. This information, if available at all, can usually be gleaned from the labors of taxonomists.

An excellent example of the importance of taxonomy is provided by recent efforts to establish the Australian hydrilla stem weevil (*Bagous hydrillae*) at sites in the United States. Characteristics of Choke Canyon Reservoir in southern Texas caused us to believe

that this was an ideal location for this insect. Although *B. hydrillae* lays eggs in submersed hydrilla and the larvae can develop underwater in the stems, very few pupae develop unless plant material containing fully grown larvae becomes stranded on dry soil. Pupae are then formed among the dry hydrilla strands or in the soil below. Water levels had progressively receded at Choke Canyon over several months. As a result, large amounts of hydrilla had become exposed on the drying hydrosols. Also, wind and wave action on the lake caused the plant material to fragment and drift on shore creating an extensive strand line composed mostly of hydrilla. These two conditions appeared to provide abundant habitat for this insect as well as ideal pupational sites.

We cooperated with Dr. Grodowitz and Dr. Cofrancesco of WES and with the Texas Parks and Wildlife in a concerted effort to release and establish *B. hydrilla* at the Choke Canyon site during 1994. Our efforts were rewarded very quickly when large numbers of weevils began appearing in samples collected by personnel from WES, whereas they had never found *any* weevils in samples collected before the release of *B. hydrillae* at the site (Grodowitz, pers. comm.). We subsequently also reared them directly from hydrilla collected at the site, confirming that they were, in fact, utilizing hydrilla as a host. This led us to believe that a *B. hydrillae* population had established. Two actions were therefore considered: (1) discontinue the release program at that site to determine if the population could persist and (2) discontinue the time-consuming and laborious process of mass rearing weevils, assuming that we would soon be able to easily acquire them from the field for future dissemination. Fortunately, however, before implementing either of these actions we provided specimens to Dr. C. W. O'Brien, a taxonomic expert on the genus *Bagous*. He determined that the Choke Canyon weevils were not *B. hydrillae*, but rather *B. restrictus*, a native species. *B. restrictus* closely resembles *B. hydrillae* but was not previously known to utilize hydrilla as a host, despite the fact that we were able to rear them on it. The availability of this taxonomic expertise provided the information that we needed to con-

tinue rearing and releasing *B. hydrillae*. As a result of continuing to release at the site, *B. hydrillae* now (Jan. 1995) truly seems to be established at Choke Canyon.

Assessment of effectiveness of biological control agents after their release is another important type of foundational research. This aspect of the program has no direct bearing on the success or failure of a particular biological control agent and is quite expensive and time consuming to carry out. As a result, funds are usually allocated toward more critical aspects of the project and minimal effort is usually directed toward assessing efficacy. However, several good reasons exist for evaluating biological control projects: (1) The introduction of one particular biological control agent before another is often based on a priori predictions of their relative effectiveness. It's therefore valuable to know if these predictions were accurate. Effectiveness is generally the least predictable characteristic of a particular agent. If the most effective agents could readily be picked from among an array of candidates, progress on biological control projects could be accelerated; (2) As mentioned in the above example on the Australian mimosa project, evaluations provide valuable feedback, particularly to foreign aspects of the research, to ascertain whether more agents will be needed and what characteristic they should possess; (3) It's also important to advertise success in biological control projects for the dual purposes of ensuring future financial support and for garnering favorable public opinion. Favorable public recognition is one of the primary reasons that many sponsoring agencies support biological control (Harris 1989). It's often difficult to ascertain the success or failure of a project without a proper evaluation. This is especially true if the effects of the biological control agents are subtle and not readily evident to the casual observer; (4) Considerable funds are often spent to redistribute a biological control agent after its initial release. It's a waste of money to do this unless the agent is of proven value; and lastly, (5) we have a scientific obligation to evaluate the effects of releasing a new organism into the environment. The introduction of a plant-feeding insect, especially one capable

of devastating an invasive plant species that dominates large areas, potentially has broad, regional (hopefully beneficial) effects. Ecologists are now beginning to recognize this and are expressing concern (e.g., see Simberloff 1992). Hence, delineation of these effects should be part and parcel of all biological control attempts.

Unfortunately, it is not always easy to evaluate the performance of a particular biological control agent or the results of a complete biological control program. The released biological control agents might need a quite long period of acclimatization before they become abundant. In fact, Harris (1989) noted that this acclimatization can require up to 15 years before the biological control agents adapt to local conditions. Evaluations done too early can produce erroneous conclusions. When effects are rapid and readily observable, evaluations scarcely need to be done. However, effects are more likely to be subtle and result in long-term declines or cause the target to be susceptible to secondary stresses such as disease, drought stress, or temperature extremes.

Although one might easily assume that monitoring is worthwhile, this is not necessarily the case. Dilettantes typically call for "monitoring" of biological control agents and the target weed even before the release of an agent so as to establish baseline data from which to measure changes. Such monitoring usually involves a "shotgun" approach to data collection entailing measurement of as many parameters as possible (often excluding the few that eventually are found to matter). This requires tremendous amounts of time and effort. Monitoring of a single agent could require 5-10 scientist-years (SYs), whereas an entire biological control project can usually be accomplished in 20 SYs (Harris 1989). Further, monitoring can usually be done at only a few locations (perhaps the wrong ones) and therefore fails to provide broad regional information. In addition, it usually lacks controls and therefore does not provide scientifically acceptable proof of the effects of the agents. The effects of an agent as measured by deviations from baseline conditions are difficult to separate from natural changes re-

sulting from plant succession, climatic trends, or other long-term environmental changes. Instead of wasting time and resources on "monitoring," it is much more appropriate, when possible, to conduct field experiments utilizing appropriate controls and to focus on answering specific questions. The most important part then becomes the formulation of appropriate questions and the design of efficient experiments. This requires preliminary, smaller scale studies conducted in tanks or perhaps pools. Field experiments can then be devised to determine the general applicability of the results. However, uninitiated observers often do not recognize the need or importance of such experimentation and often criticize this activity as having dubious applicability to field conditions.

Our experience in evaluating the effects of biological control agents on waterhyacinth populations should serve as a valuable object lesson for the above points. Initially, beginning in the early 1970s, we monitored several waterhyacinth populations to determine the effects of *Neochetina eichhorniae*, *N. bruchi*, and *Sameodes albiguttalis*. We successfully documented the decline of waterhyacinth at a north Florida lake after first developing baseline data (cf. Center and Spencer 1981). The decline occurred over a 4-year period and the associated study required nearly 8 years of field work (Center, Cofrancesco, and Balciunas 1990). In addition, a number of observers subsequently documented waterhyacinth declines induced by these insects both in the United States and in other countries (Beshir and Bennett 1985; Bodle 1988; Center 1987; Center and Durden 1984, 1986; Center, Cofrancesco, and Balciunas 1990; Cofrancesco 1985; Cofrancesco, Stewart, and Sanders 1985; DeLoach and Cordo 1983; Girling 1983; Goyer and Stark 1981, 1984; Haag and Center 1988; Jayanth 1987, 1988; Wright 1979, 1981). However, skeptics dismiss all of this as anecdotal evidence and invoke other causes to explain these declines (however, cf. Center 1987). As a result, biological control agents, as resources for waterhyacinth management, have not received the attention they warrant, particularly as components in integrated management schemes.

In an attempt to address this lack of proof of field efficacy, we conducted two studies that were sponsored by the Florida Bureau of Aquatic Plant Control, Department of Environmental Protection (DEP). In one study, we investigated the compatibility of traditional waterhyacinth maintenance procedures and biological control. We've long noted the paucity of biological control agents at sites where the resident waterhyacinth populations are subjected to periodic herbicidal treatment, and we've suspected that this was due to incompatibility between herbicidal control and biological control. We therefore compared weevil populations, as well as the status of the waterhyacinth populations, between maintained and unmanaged sites. Samples were collected at a total of 54 sites distributed from Tallahassee to Fort Lauderdale. The sites were selected by DEP regional biologists (RBs) from within their respective regions based on the criteria that waterhyacinth could be found in each site and that the maintenance history of each site was known.

Waterhyacinth populations at maintained sites generally tended to represent earlier phenostages (incipient or colonizing stages) and showed few effects of biological control. Also, the plants were high quality, being more succulent with higher concentrations of nitrogen in the leaf tissue. In contrast, populations at unmanaged sites tended to show high levels of stress induced by herbivores (primarily waterhyacinth weevils) and were of poor quality (e.g. lower nitrogen levels in leaf tissues). The proportion of biomass allocated to floral structures was nearly fivefold greater at managed sites than at unmanaged sites. Thus, seed production could be a much greater factor at managed sites.

Total waterhyacinth coverage was always lower at maintained sites, which averaged 15 percent as compared to 60 percent at unmanaged sites. This attests to the efficiency of maintenance programs. However, it was interesting to note that none of the unmanaged sites were completely covered by waterhyacinth, despite the lack of any control efforts over a period of several years. Prior to the introduction of biological control agents, com-

parable sites were covered bank to bank (Center, pers. obs.).

Adult weevil counts at unmanaged sites exceeded the counts at managed sites by threefold on a unit area basis and by nearly fivefold on a per plant basis. Total weevil density (larvae+adults) was about twofold greater at unmanaged sites than at managed sites (97 vs. 54 insects/m²). After factoring in the fourfold difference in waterhyacinth coverage, the differences in total weevil populations between the two groups was at least eightfold.

We also sampled the weevil populations at these sites and dissected a proportion to assess their reproductive condition. Fecundity was positively correlated with leaf nitrogen concentrations, so weevils were generally more fecund at managed sites, despite their lower numbers. This suggests that sustained weevil attack induces a reduction in plant quality (cf. Center and Van 1989) which, in turn, causes reduced fecundity of the weevil population and an easing of herbivory. These results also suggest that maintenance procedures reverse this to some degree, but, because of the concomitant loss of the herbivore population, their population response is delayed. Hence, this provides clear possibilities for integrated control using herbicides to minimize infestations and maintain plant quality while employing techniques to avoid devastating reductions of the weevil populations. The result could be longer term control at less cost.

The realization that waterhyacinth coverage was often incomplete even at unmanaged sites caused us to consider the possibility that the primary effect of the weevils was to retard rates of mat expansion. All previous studies focused on induced changes within a solid mat (usually on a per unit area basis), assuming that reductions would occur and would be observable. No one had ever examined the effects of this herbivory on rates of coverage. It would certainly be more desirable to prevent plants from infesting an area of water rather than to remove them after the area is infested. We addressed this at a site in northern Florida by creating incipient infestations of

waterhyacinth in plots that provided space for the plants to expand into and measuring the rate at which this expansion occurred. This study was also sponsored by Florida DEP.

The plots consisted of two frames, one encompassing an area of 10 m^2 the other an area of ca. 84 m^2 . The smaller frame was filled with small waterhyacinth plants and was centered within the larger frame. The inner frame was constructed of small diameter PVC-pipe that held the original plants in place but allowed them to grow over and outward. We measured area of coverage by marking grid coordinates on the outer frame which were used to map the distribution of the plants at periodic intervals. Data were also taken on the status of the plants and the condition of the weevils. Five plots were established and the inner frames were initially inoculated with 0, 1,000, 2,000, 3,000, or 4,000 weevils. These rates represented the normal range usually observed in the field. Plots were inoculated in April and data were collected at 4- to 6-week intervals over the course of one year. Plants in the control plot covered the available space very quickly, attaining 45-m^2 coverage by late July and 56 m^2 by December (a six-fold increase). The other plots ranged from 13 to 29 m^2 by late July and from 18 to 36 m^2 by December. Results were generally consistent with treatment levels, with the 3,000-weevil plot growing the least and the 1,000-weevil plot growing the most (except for the control). The exception was the 4,000-weevil plot which grew nearly as much as the 1,000-weevil plot. This was because the high infestation level caused a rapid deterioration of the plants and a responsive decrease in the weevil population. This happened early in the study, so the plants had most of the growing season to recover. Nonetheless, plant mats in all plots that were inoculated with weevils expanded more slowly than the control.

The total plant population (density per plot) increased initially in all plots, with peak numbers inversely correlated with inoculation rates. Numbers in the control plot remained steady thereafter, but populations declined in all of the plots that received weevil inocula-

tions. The original plants seemed to persist, so the plants that died were probably the younger, smaller shoots. In addition, the canopies of the plots inoculated with weevils opened up, exposing the water below, whereas complete canopy coverage was maintained in the control. This reduction in the canopy was related to reduced size of the leaves produced by the plants exposed to weevil treatments. Hence, the effect of the weevils was to cause the waterhyacinth mats to expand more slowly relative to the controls. This reduction was brought about by increased mortality and reduced plant size and leaf area (Figure 1). It was not previously known that mat expansion rate would be the most important parameter to monitor, so many earlier "monitoring" efforts were misdirected.

This study was later repeated at a second site in southern Florida with similar results. Together, the resultant information debunks many misconceptions about biological control of waterhyacinth. For example, it is commonly believed that waterhyacinth populations outgrow the weevil populations. Yet the weevil intensity (number per plant) in the controls steadily increased indicating that weevil numbers outgrew plant numbers. Also, even though the mats receiving the highest weevil inoculations only doubled over the 8-month period, they did increase. Obviously, if this occurs at field sites over a period of years with no control measures being taken, waterhyacinth infestations will eventually cause problems, even when the plants are exposed to intense herbivory and the mat expansion rate is slow.

This study clearly shows that, even though present biological control agents serve a useful function, other controls are needed. The options for providing better biological control are: (1) to manage the populations of biological control agents presently available or (2) introduce new agents. The options for introducing new agents include (1) further developing previously known but incompletely studied biological control agents from South America or (2) find new, previously unknown agents.

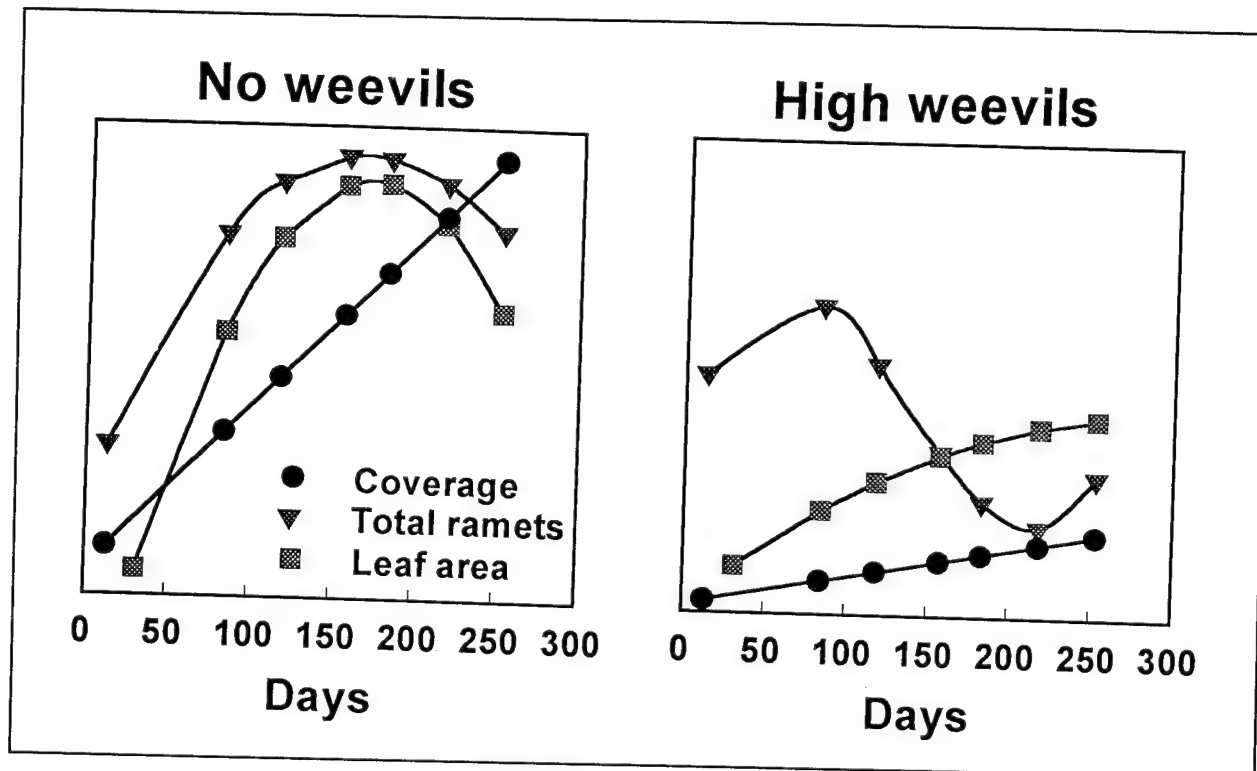


Figure 1. Partial results of a study conducted at the St. Marks River in northern Florida which compared the effects of inoculating 10-m² waterhyacinth mats with high levels of waterhyacinth weevils (300 weevils/m²), initially vs. no initial inoculation of weevils. The data show waterhyacinth population growth (total ramets per plot), plant size (area per leaf), and rates of mat expansion (coverage)

The Jacksonville District has encouraged the development and introduction of new agents. In response, we are now arranging for the South American pyralid moth *Acigona infusella* to be tested in Australia to determine if it is a suitable candidate for introduction into the U.S. This determination will be made based upon the extent of the threat that it might impose to pickerelweed (*Pontederia cordata*). Silveira-Guido (1965, 1971) considered *A. infusella* (Walker) to be one of the most important phytophagous species on waterhyacinth. It has never been reported from hosts other than members of the Pontederiaceae in natural conditions. We are concerned, though, that *A. infusella* might cause serious damage to *P. cordata*. Silveira-Guido (1965) intimated that *A. infusella* was not as common on *P. cordata* as on *E. crassipes* and *E. azurea*. His data were, unfortunately, contradicted by DeLoach et al. (1980). They found *A. infusella* to be occasionally abundant on *P. cordata* in natural conditions, sometimes more so than on *E. crassipes*.

We have not previously recommended *A. infusella* for further study because of the possible importance of *P. cordata* to wildlife in the United States and its status as a native. If, however, it was found that field populations of *P. cordata* are not severely damaged by this insect, then the matter might be reconsidered. We discussed the possibility of initiating this work with scientists from the CSIRO Long Pocket Laboratories in Indooroopilly, Queensland. They were quite willing to undertake the project if funds could be obtained to support it. Because they already have permission to release *A. infusella*, it would only be necessary to establish field populations in areas where pickerelweed occurs and evaluate any impacts on it. The Jacksonville District has now funded this work and, with some support from FDEP, the project is now underway. This is a clear example of the use of foundational research to overcome an obstacle in the development of a potential biological control agent.

In summary, foundational research is vitally important to biological control. Dr. Lloyd Andres (1981) perhaps said it best: "Although biological control has provided practical solutions to problems and promises to continue to do so, it is the information gathered in the problem-solving process that makes biological control such an important part of any pest management program."

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Hydrilla Defense Genes: Implications in Biocontrol

by
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Background

The 25th Proceedings of the Aquatic Plant Control Research Program entertained discussions on biotechnical approaches to aquatic plant management. Those discussions centered on genetic engineering (Kees and Theriot 1991) and allelopathy (Jones and Kees 1991). The next step was to determine the feasibility of applying molecular biology techniques to aquatic plant biocontrol research, specifically, and to aquatic plant control research, in general. The capability for molecular genetic analysis constitutes a new technical capability for WES and in an effort to develop this technology area, advanced training in molecular biology was acquired during a 16-month study in residence at Purdue University. The first project at WES designed to apply this technology to aquatic plant biocontrol research was funded in 1992 through the WES Director's Laboratory Discretionary Research and Development Program and is scheduled for completion in FY95; some of the preliminary findings are reported here. Also, some potential applications of this technology to aquatic plant biocontrol research and some potential benefits to other aquatic plant control research technologies are discussed briefly.

Introduction

Plants exhibit natural resistance to disease including inducible plant defense mechanisms such as the accumulation of phytoalexins, the formation of protective structures such as papillae, and increases in the activity of certain hydrolytic enzymes (Sequeira 1983). An increasing number of researchers of terrestrial plant-pathogen interactions have found it possible to establish a direct correlation between

the timing of certain genetic events and the onset of a quantitative physiological defense response, of a phenolic nature, in the host plant (Bell et al. 1986; Lawton and Lamb 1987). These data show that specific accumulation of plant defense gene transcripts is a key early component of defense responses in wounded plant tissue (such as might occur during insect feeding) as well as in fungally challenged plant tissue. There is also conclusive evidence that additional phytoalexin synthesis occurs in surrounding cells (Snyder and Nicholson 1990) and in distant, unelicited tissue (Lawton and Lamb 1987). The gene transcripts in question code for key enzymes in phenolic compound biosynthesis (i.e., enzymes of the shikimic acid pathway). Many of the phytoalexins that have been implicated in plant defense are either members of the group known as phenylpropanoids or members of the flavonoid/isoflavonoid group. Phenylalanine ammonia lyase (PAL) is the first committed enzyme in phenylpropanoid biosynthesis and Chalcone synthase (CHS) is the first enzyme in the flavonoid/isoflavonoid branch of this pathway. Investigators analyzing the phenolic content of hydrilla grown in culture have reported that the major compound in both leaves and stems is a caffeic acid ester which constituted as much as 0.273 percent of the leaf fresh weight (Hipskind et al. 1992). Caffeic acid is the first metabolic intermediate of the phenylpropanoid pathway. If hydrilla utilizes this compound as a primary defense against fungal pathogens and/or for de novo synthesis of other fungitoxic phenols, this pathway could prove to be of critical importance in the initial and long-range efficacy of microbial biocontrol agents. A temporal analysis of early genetic events correlated with the appearance and type of phenolic products will tell us how quickly hydrilla activates this

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defense mechanism and the relative fungitoxicity of both residual and induced phenolic species. This information can then be used to enhance evaluation of potential mycoherbicides and to improve the efforts of integrated approaches that use some combination of existing aquatic plant control technologies.

Research Objectives

The general objectives of this study are to (1) establish a model for genetic analysis of the interaction between an aquatic plant and a fungal pathogen and (2) identify potential applications of this technology area to aquatic plant control research. The specific objectives of this research are to (1) show that hydrilla has the genetic machinery for two important enzymes in phenolic compound biosynthesis, (2) demonstrate that the active gene transcripts for these two enzymes increase in response to fungal challenge, (3) identify the most abundant residual and induced phenolic compounds, and (4) evaluate the fungitoxic effect of these compounds on the growth of selected fungi.

Research Approach

Algal-free dioecious *Hydrilla verticillata* serves as the host plant for this plant-pathogen model; *Mycoleptodiscus terrestris* serves as the pathogen. Hydrilla leaf and stem tissues are being used for genomic DNA extraction and cDNA gene clones from barley and sorghum serve as heterologous probes for PAL and CHS, respectively. Hydrilla tissues are being genetically analyzed over eight discrete time intervals after pathogen exposure for the presence and quantity of PAL and CHS gene transcripts (messenger RNA). High pressure liquid chromatography (HPLC) is being used to identify dominant phenolic species that appear in hydrilla at these same time intervals. Once these compounds have been identified, they will be evaluated in laboratory bioassays at physiological concentration to determine their ability to inhibit radial growth of selected fungi in plate culture. Light microscopy studies are underway to establish the timing of important biological events in the fungus such as

spore germination and appressorium (infection structure) formation.

Research Results

When this research was initiated in 1992, an extensive literature search produced no published accounts of DNA isolation from hydrilla tissues. After a number of unsuccessful attempts to isolate hydrilla genomic DNA, a protocol for isolation of DNA from rice reported by Dellaporta, Wood, and Hicks (1983) was successfully adapted to hydrilla tissues. Preliminary gene screening studies (Southern Hybridization Studies) suggest that genes for the two enzymes of interest are present in the hydrilla genome. We are in the process of determining whether each gene exists singly or as a gene family. We have also been unsuccessful in locating literature citations for the isolation of either total RNA or poly A⁺ messenger RNA (mRNA) from hydrilla tissues. We continue to evaluate a number of protocols and modifications to that end.

Preliminary HPLC results from methanol-extracted hydrilla leaf tissue appear to indicate the presence of three major peaks that rise and fall temporally after pathogen exposure. When these peaks are identified, fungitoxicity studies will be initiated.

Light microscopy studies have also been initiated.

Implications in Aquatic Plant Biocontrol Research

Our interest in this technology area in 1991 was basically to determine whether molecular biology techniques could be usefully applied to aquatic plant biocontrol research. Today we can answer that question with an enthusiastic "Yes!"

As reported by Theriot, Cofrancesco, and Shearer (1993) questions were raised regarding the identity of the [hydrilla pathogen] *Macrophomina phaseolina* (M.p.) which was later confirmed to be *Mycoleptodiscus terrestris* (M.t.) by B.C. Sutton at the International

Commonwealth Institute (Shearer 1994). Shearer provided several isolates of *M.t.* and several isolates of other fungi that resemble *M.t.* in culture to be used in a blind DNA fingerprinting study to see if these two genera could be distinguished. A preliminary DNA fingerprinting study which uses randomly amplified polymorphic DNAs (RAPDs) was able to resolve differences not only between the two genera, but also within species of the same genus. With the advent of this technology coming online at WES, we now have the capability to environmentally track any microbe being evaluated as a biocontrol agent in a consistent and unambiguous manner. Likewise, DNA fingerprints can be developed for target aquatic plants to expose variations in biotype which might significantly influence plant nutritional quality. If a particular genetic pattern can be linked to a particular biocontrol agent response, these findings could have far-reaching implications in biocontrol insect release and establishment efforts and pathogen selection and evaluation regimes.

Implications in Aquatic Plant Control Research

Several promising applications of molecular biology techniques to aquatic plant biocontrol research have been identified primarily because a specific need was articulated. Application of this technology to aquatic plant control research will probably require a similar articulation of need. But even now, ecologically speaking, these techniques can be used to study the genetic basis of aquatic plant invasions and declines; chemically, we can begin to investigate the genetic basis for variations in aquatic plant response to chemical treatment; and our simulation technology will benefit from advances made in biocontrol, ecological, and chemical technology areas. We can say with relative certainty, however, that our current vision of potential research applications for this technology, at best, represents only the "Tip of the iceberg."

Future Directions

Heightened environmental awareness has precipitated the need for the development of more environmentally sensitive solutions to environmental problems. Our insect biological control agents have addressed this challenge well, but there are instances when an insect population fails to establish at a particular site or establishes poorly. It would be useful to know if several biotypes of a target plant are present in the same community, whether perceived genetic differences can be linked to insect feeding preferences, etc.; DNA fingerprinting studies can provide this kind of information. A group in Minnesota and a group in Florida have already initiated DNA fingerprinting studies on *Myriophyllum spicatum* and *Hydrilla verticillata*, respectively. A similar effort should be engaged for *Eichhornia crassipes* and other target aquatic plants. As we continue to develop mycoherbicides for aquatic plant management it will become necessary to be able to environmentally track these organisms and distinguish between isolates of the same species; again, DNA fingerprinting studies can render this capability. The technology for genetic manipulation (i.e., alteration) of bacteria and fungi is also available and represents yet another option for the development of microbial biocontrol agents.

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Foreign Surveys and Quarantine Studies for Biocontrol of Eurasian Watermilfoil and Hydrilla

by
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This year we continued the studies reported at last year's Aquatic Plant Control Research Program (APCRP) meeting (Bennett 1994, Buckingham 1994). These included surveys in the People's Republic of China for insects on Eurasian watermilfoil (*Myriophyllum spicatum* L.) and hydrilla (*Hydrilla verticillata* (L.f.) Royle) and quarantine studies in Florida with insects collected during the surveys. New this year was a plant pathogen survey conducted in China along with the insect survey. We had a major turnover in personnel this year, with Ms. Christine Bennett, Mr. Jason Carter, and Ms. Yanfen Chen working on the project at the end of the year.

Foreign Surveys

Our foreign program continues in cooperation with the Sino-American Biological Control Laboratory (SABCL), Biological Control Institute, Chinese Academy of Agricultural Sciences, Beijing. Mr. Zhiquan Chen, who joined our aquatic weed program in 1992, collects insects, surveys, and accompanies us during our field surveys. This year he returned in June to Shenyang, Liaoning Province, to look for cocoons and adults of a donaciine leaf beetle. This beetle was collected as larvae on the crowns and lower stems of unhealthy looking hydrilla in 1992 (Buckingham 1993). Mr. Chen found very little hydrilla and no insects were found on it. However, two adults of a small species of *Macrolea*, a donaciine leaf beetle genus, were collected sitting on leaves of waterchestnut, *Trapa* sp., at the site. *Macrolea* includes species reported from watermilfoils and thus there is a good possibility that these adults are the target species. Three larvae collected in 1993 (Bennett 1994)

did not survive the winter in the SABCL greenhouse. In July, Mr. Chen returned to Shenyang with Willey Durden, USDA/ARS, Fort Lauderdale, FL, to again search for these leaf beetles. As in June and in 1993, there was little hydrilla in the roadside canal that had been heavily infested in 1992 (Buckingham 1993). However, small numbers of medium-sized larvae were found on the lower stems. These larvae were carried to SABCL and were fed hydrilla in moist containers. They made small feeding holes in the stems. None lived to maturity, so we are still unsure of their identity.

Dr. Judy Shearer, plant pathologist, U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS, joined Mr. Durden and Mr. Chen in mid-July to accompany them on surveys of hydrilla and Eurasian watermilfoil in the surroundings of Beijing (Shearer 1995).

Adults of a *Bagous* weevil that fed and oviposited in cages on hydrilla were collected in a small drainage pool, but no larvae were found in hydrilla stems in the field. Instead, they attacked the hydrilla relative *Hydrocharis dubia* (Bl.) Backer. Studies were stopped when it was determined that larvae did not develop on hydrilla and when the true host plant was determined.

I arrived in Beijing on 6 August with a shipment of waterhyacinth weevils, *Neochetina bruchi* Warner, from Dr. Ted Center, USDA/ARS, Fort Lauderdale, FL. The weevils were requested by Dr. Lu Qing Guang, Director of SABCL, for quarantine studies to determine their safety for eventual release in south China. The Biological Control Institute maintains a quarantine laboratory down the hallway from the SABCL offices.

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On 8 August, Mr. Chen and I departed for Harbin, Heilongjiang Province, to collect weevils on Eurasian watermilfoil. Our local host again this year was Mr. Wang of the Plant Protection Institute. High water at our Shi-er-li-Pao marsh site (150 km west of Harbin) (Bennett 1994, Buckingham 1993) prevented wading and sampling along transects as planned. High water and floods had also closed the highways, which prevented a planned survey near Jiamusi and Fujin in the northeast corner of Heilongjiang Province. We collected plants and weevils at Site 1 along the eastern shore of the marsh by sitting on a luggage belt crisscrossed on an automobile innertube. We were able to maneuver through the mats to collect but not to adequately sample a transect. At Site 2 along the southern shore of the marsh at a small bridge, we rented a pole boat. Maneuvering in the swift water rushing from the marsh to the bridge was very difficult. Thus, we were limited to collecting from a stationary position along a cattail island about 50 m from shore.

The watermilfoil at Shi-er-li-Pao marsh was all *Myriophyllum verticillatum* L. No *M. spicatum* was found at the two sites, although it was the dominant species in 1992 and might have been present in 1993. Larvae, pupae, and teneral adults (soft, newly emerged) of our target *Bagous* sp. weevil were collected in tunnels in the damaged stems. Adult *Eubrychius* sp. weevils that feed underwater on watermilfoil leaves were collected at both sites but were exceptionally heavy at the cattail island, Site 2. They were observed at noon climbing from the water onto the boat, emerged plant parts, flotsam, algae, etc. None were observed flying, but several were observed extending their wings to dry as they emerged from the water. This suggested that they might be migratory. I watched a couple of them for several minutes each as they emerged, but they remained stationary as they dried.

We spent our limited time collecting rather than observing. The water beneath the flotsam was checked for watermilfoil; but only a narrow-leaved species of *Potamogeton* was present, and it was not attacked. Neither were

the watermilfoil plants in the surrounding area. It appeared that these weevils had ridden on floating watermilfoil stems to the island and had emerged seemingly in response to the noon sunshine. If the small island had not disrupted their journey, the stems would have floated to the nearby shore or out of the marsh. These adults might have been headed for winter diapause. The numbers crawling from the water seemed to decrease about 1:30 p.m. when shadows covered the search area and we stopped observations. A decrease, however, might have been due to our heavy collecting rather than to lack of sunshine as it appeared to be.

Nine *Bagous* sp. adults from watermilfoil were collected sitting out of water with the *Eubrychius* sp. Over half of a sample of 20 watermilfoil roots at this site had donaciine (probably *Macrolea*) larvae attached. They were also found at Site 1. *Phytobius* sp. weevils were found at Site 1, but numbers were small. Almost all flower stalks in one area were damaged, many with no seeds; however, immatures, including empty cocoons, were rare.

Close to QiQiHar, west of Shi-er-li-Pao, we found *M. spicatum*, *M. verticillatum*, and hydrilla together at a reservoir and surrounding drainage areas at Qi Lin Dao Qun. At one site a few *Phytobius* sp. adults and larvae were found on *M. verticillatum* but not on *M. spicatum*. A short distance away, cocoons were found on both plant species but mostly on *M. spicatum*. This suggested that perhaps two species were involved, and indeed, two species were found in the weevils mixed from all sites and carried to quarantine. We do not know if the two species have different host ranges. Donaciine larvae and cocoons were collected on roots and lower stems of both species of watermilfoils, hydrilla, *Nymphaea tetragona*, *Potamogeton lucens*, *P. perfoliatus*, *Nymphoides peltata*, and an unknown grass or sedge. *Macrolea* adults, which have not yet been identified, were dissected from some cocoons. I am unsure if there is one polyphagous species or if multiple species are involved.

Hydrilla formed small dense mats at these sites but only in competition with the other species that also formed dense mats. *Myriophyllum spicatum* was the dominant species at one drainage site and *M. verticillatum* at another. No species was dominant along the shore of the reservoir where many plants appeared to have floated in from deeper water, although hydrilla was most abundant.

Two additional sites were surveyed. Long An Qiao, a marsh to the north of Qi Lin Dao Chun and northeast of QiQiHar, had both species of watermilfoil and hydrilla. *Eubrychius* sp., *Phytobius* spp., and possibly *Bagous* sp. were collected in small numbers on the watermilfoils. Northwest of Harbin along the west side of the Songhua River toward Chang Fa, a few *Phytobius* sp. cocoons were collected on *M. spicatum* in a small lake, but no *Bagous* sp. were found. In a drainage channel along the highway, *Phytobius* sp., *Eubrychius* sp., and *Bagous* sp. were collected on *M. verticillatum*. No *M. spicatum* was present. *Phytobius* sp. cocoons were very abundant, and most of the adults carried to quarantine came from the cocoons at this site. The cocoons were heavily parasitized by small wasps both here and at other sites.

A total of 113 *Bagous* sp. adults, 97 *Phytobius* spp. adults, and 49 *Eubrychius* sp. adults were carried to quarantine at the end of August.

Quarantine Studies

Watermilfoil stem weevil

We have attempted to establish a quarantine colony of *Bagous* sp. for three seasons (Bennett 1993, Buckingham 1994). We were able to obtain a few F_3 adults in 1992 and a few F_2 adults in 1993 before the adults stopped reproducing and apparently entered winter diapause (resting stage). In January 1994, 25 F_1 adults were placed in a temperature cabinet that was slowly cooled to 7 °C daytime (10 hr) and 2 °C nighttime (14 hr). Half were removed after 17 days and slowly warmed to constant 24 °C (16 hr light). The second

group was removed to the same conditions after 58 days. Nine parents were held for 27 days in the cabinet at 7 °C before removal to 24 °C.

After a month at 24 °C, four females from the first F_1 s removed were placed in a greenhouse tank with potted milfoil. Two months later, two dead mature larvae were found in the milfoil, indicating that diapause had been broken in at least one female. At the same time a dead pupa was found in planted milfoil in a gallon jar with the parents. These had been held at 24 °C in the cabinet. Although the progeny in these experiments had died, probably from lack of air in waterlogged stems, diapause (in at least some adults) had been broken by relatively short periods of cold (17 and 27 days).

The adults carried to quarantine in August 1995 are continuing to produce progeny under 16 hr light. Thus, none have been refrigerated. Production is light, suggesting that some are in diapause, but we will continue to rear them until oviposition stops.

Host range testing has been delayed until we have a colony and achieve consistent feeding and oviposition results with our watermilfoil controls. However, we have exposed a few species along with *M. spicatum* in our rearing cages (plastic refrigerator boxes with moist toweling). There was no feeding on *Cabomba caroliniana* Gray, *Hydrilla verticillata* (L.f.) Royle, *Najas guadalupensis* (Spreng.) Magnus, *Potamogeton amplifolius* Tuckerm., and *Ranunculus* sp., and minor to moderate feeding on *Myriophyllum aquaticum* (Vell.) Verdc., *M. heterophyllum* Michx., and *M. sibiricum* Komarov.

Watermilfoil flower weevil

Initial biology and host range studies with *Phytobius* sp. were reported last year (Buckingham 1994). These could not be reared continuously in quarantine because watermilfoil stopped flowering in November. This year, two species were found mixed in the adults carried to quarantine. The species studied last year had a slightly longer snout than the

new one. Because the short-snout species had not yet been studied, it was chosen for initial studies. However, the colony was lost when watermilfoil stopped flowering. We are attempting to rear the long-snout species with nonflowering watermilfoil.

In development studies, the short-snout species deposited viable eggs, 96 percent ($n = 104$), on *M. spicatum* flower stalks, but only six cocoons were formed and four adults emerged (1f,3m). Development time was 13 to 14 days (27°C), the same as that of the long-snout species (Buckingham 1994). Two new females deposited totals of 66 and 91 eggs before stopping oviposition. The eggs were placed in feeding scars on the flower stalk hidden by female flowers and among the male flower buds. Adult feeding was primarily on the flowers, but the flower stalks were also eaten. The biology of this species appears very similar to that of the long-snout except for the poor larval survival. Perhaps *M. spicatum* is not a good host (most adults collected this year were on *M. verticillatum*) or perhaps our rearing procedures were inadequate for this species.

Some host range tests in addition to those reported in Buckingham (1994) were conducted with the long-snout species at the end of last year and with the new adults at the end of this year (October-December). There was no feeding on *Rumex verticillatus* L., only nibbling on *Polygonum hydropiperoides* Michx., and moderate to heavy stem and leaf feeding (compared to *M. spicatum* without flowers) on *Proserpinaca pectinata* Lam., *P. palustris* L., and *Myriophyllum aquaticum*. A few eggs were also deposited on the latter three species. Additional tests will be conducted when a colony has been established.

Milfoil leaf weevil

We were unable to do much with *Eubrychius* sp., an underwater relative of the *Phytobius* spp. flower weevils. Eggs were laid on leaves at the tip. Both adults and larvae ate the tips and surrounding leaves. A flower bud was also damaged, and the stem of a side shoot was cut. Pieces of leaves and stem floated in

the cages. Spherical parchment-like cocoons were embedded in the stems, thus appearing hemispherical like the *Phytobius* cocoons. They were smaller and darker than the light brown *Phytobius* cocoons, almost black. Stems were often deformed at the cocoon. Not many larvae were produced, but most of those died even though the water was aerated. This might indicate that *M. spicatum* is an inadequate host, although the rearing technique is also suspect.

In one small 7-day multichoice adult feeding test, there was no feeding on *Potamogeton amplifolius*, *P. crispus* L., *P. foliosus* Raf., *P. nodosus* Poir., and *Ranunculus* sp. Feeding, although much less than on *M. spicatum*, was found on *M. heterophyllum* and *M. sibiricum*. *Eubrychius* sp. adults were also collected on *M. verticillatum* in Xinjiang Province in western P.R. China in 1991 and carried to quarantine (Buckingham 1992). Those adults appear to differ from these, but they have not yet been compared by Dr. O'Brien, Florida A&M University.

Future Plans

In summer 1995, additional surveys for insects and pathogens on watermilfoils and hydrilla will be made in P.R. China. Possibly initial contacts will be made in Russia for information on insect herbivores of the target plants. Additional collections of *Bagous* sp., *Phytobius* sp., and *Eubrychius* sp. on watermilfoils will be made near Harbin in Heilongjiang Province. Populations on both species of watermilfoils will be kept separate. An attempt will again be made to collect adults of the donaciine leaf beetle, possibly *Macrolea* sp., on hydrilla at Shenyang, Liaoning Province. Biology and host range studies will be conducted in quarantine with the various species, if we can establish colonies. Mr. Chen Zhiquan, SABCL, will again travel to Gainesville during the winter for training.

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The Use of Pathogens for the Management of Hydrilla and Eurasian Watermilfoil

by
Judy F. Shearer¹

Introduction

When the pathogen biocontrol program was initiated the focus was on discovering and developing endemic pathogens for management of hydrilla and Eurasian watermilfoil. This approach limits the magnitude of the survey for pathogens to sites of hydrilla and watermilfoil infestation within the United States. Potential biocontrol agents are obtained from the target plants by plating stem and leaf tissue onto selective media and isolating microbes which grow out from the plant pieces. Species known to have pathogenic properties undergo a screening procedure to identify those isolates which are efficacious on the target plant. Isolates which show promise undergo additional testing in small-scale laboratory and field studies. It is the intent that an agent obtained in this manner will be used to augment the naturally occurring microbial population and by using high enough dosage rates the targeted plant can be reduced to a manageable level. For ease of use, the agent must be formulated so it can be applied in much the same manner as a herbicide. This approach using plant pathogens is known as the inundative method of biocontrol (Templeton, TeBeest, and Smith 1979). A strain of *Mycoleptodiscus terrestris* (Gerd.) Ostazeski from Texas has been identified as a potential agent for hydrilla management and although a strain of *M. terrestris* from Massachusetts has shown some promise for milfoil management searches continue for additional agents.

Only recently has the pathogen program been expanded to include foreign exploration for pathogenic microorganisms of hydrilla and Eurasian watermilfoil from their native

ranges. The focus for this approach will be to find organisms which can be used as classical biocontrol agents.

Endemic Studies

Hydrilla verticillata

One of the main efforts in the hydrilla work unit during 1994 was to begin formulation of *M. terrestris*. The development is a formidable task because the aquatic environment presents a new medium for bioherbicide formulation research. Development of formulations for use on terrestrial weedy species has taken years and, while the knowledge gained is of great value, the designs are not readily adaptable for use in aquatic systems. For example, moisture limitation (i.e. a wetting time for initiation of fungal growth and invasion), a major problem in terrestrial bioherbicide design, is not a problem in aquatic systems. On the other hand, aquatic bioherbicide design must allow for dilution factors and water movement.

A contractual agreement was initiated in August 1994 with Ricerca, Inc., Painesville, OH, to design a formulation of *M. terrestris* which would be effective in an aquatic environment. To accomplish such a task three areas of research must be given consideration: (1) optimizing fermentation of the biological agent; (2) development of a compatible carrier which will adhere to the plant; and (3) incorporation of the biological agent into the carrier.

Previous studies on dosage response of *M. terrestris* on hydrilla planted in small tanks produced dramatic results within 7 days at low, medium, and high dosage rates.

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Hydrilla leaves became chlorotic and stems readily fragmented. However, if the treatment did not result in good coverage on the plant, even at high dosage rates, hydrilla was able to regrow. A more effective level of control might be achieved by treating plants with a second application of the fungus before regrowth and while the plants are under stress from the first application. To test this hypothesis tanks were planted to hydrilla and a second treatment of *M. terrestris* was applied to randomly selected tanks 2 weeks after the initial application.

One of the main considerations in management of aquatic plants is determining the best time to treat the target plant to achieve the desired results. When utilizing pathogens as biocontrol agents, phenology of the plant is of major importance. Pathogenic organisms affect plants differently depending on their stage of growth. For example, organisms which cause damping off disease affect seedlings when plant cell walls are thin, but have little impact on older plants once the walls become thickened. To determine the best time to apply *M. terrestris* in a field situation, applications were made to hydrilla at monthly intervals during the growing season.

Myriophyllum spicatum

Results of field tests using *M. terrestris* on Eurasian watermilfoil during 1992 and 1993 were disappointing. While excellent control was documented in laboratory and greenhouse trials, comparable results were not achieved in field applications. It was determined that a reassessment of endemic organisms which might have potential as biocontrol agents for milfoil be undertaken. This was possible because of the extensive culture collection which has accumulated over the past few years in the biocontrol laboratory. As part of our standard operating procedures, isolations are routinely made from plant material either sent to the laboratory or collected as part of ongoing research.

A mass screening of isolates obtained from milfoil from various regions of the United States was initiated in the fall of 1993. From

the screening, six candidates were identified for additional testing. From these six isolates, three were chosen based on efficacy results in the laboratory to be field tested at the LAERF, in Lewisville, TX.

Materials and Methods

Cultures of *M. terrestris* were plated onto Potato Dextrose Agar (PDA) (Difco Laboratories, Detroit, MI) and grown in the dark at 28 °C for 7 days. Mycelial disks were cut from the colony edge and placed in 1-L Erlenmeyer flasks half filled with Richard's V-8 broth. The flasks were shaken on a gyrotory platform shaker (New Brunswick, Edison, NJ) at 200 rpm for 6 days. The mycelial mat which developed in the flasks was filtered through four layers of cheesecloth and ground in a blender with a small amount of sterile water for 30 sec or directly ground without filtering. The resulting slurry was used as the test inoculum in aquaria, tanks, or field plots. Colony-forming unit (CFU) counts were determined by dilution plating.

Field plots approximately 1 m by 1 m by 1 m were set up in hydrilla- and milfoil-planted ponds at LAERF. Each plot was enclosed in plastic to keep the liquid inoculum within the boundaries of the plot. Randomly selected hydrilla plots were inoculated monthly starting in June. A fungal application was made to the milfoil plots in early June, but because there was a problem with the attachment of the plastic, plots were relocated and inoculated in early August.

Dosage rates were applied to the target plants as follows: 50-L aquaria received 20 ml of inoculum; 2,200-L tanks received 1,000 ml of inoculum; field plots of hydrilla received 1,000 ml of filtered inoculum; field plots of milfoil received 2,000 ml of unfiltered inoculum.

The inoculum was applied to the water surface and allowed to dissipate naturally through the plant mat. Plants were harvested from aquaria 2 weeks post inoculation and from tanks and field plots 1 month post inoculation. Aboveground biomass was collected

by clipping the plants at the sediment surface. The plants were washed to remove surface debris, dried at 60 °C for 5 days, and weighed.

Results and Discussion

Hydrilla verticillata

Preliminary screenings of *M. terrestris* with the multitude of possible carrier compounds turned up very few incompatibilities (i.e., the compounds were not lethal to the fungus). Formulations being researched but still in their early developmental stages are extruded granules, suspension concentrates, and inverts. Simple techniques such as milling following fermentation were found to substantially increase the number of CFUs. Compared to unmilled counts, milling in a blender for 160 sec produced an 11-fold increase in the number of CFUs and use of a tissumizer increased the count even more. Drying of the fungal mycelium, as expected, was found to reduce the viability of the fungus.

Percent reduction of hydrilla aboveground biomass did not change substantially with a second treatment with the fungus *M. terrestris* (Figure 1). Compared to dry weights of aboveground biomass of hydrilla in untreated tanks, biomass harvested from tanks receiving one versus two fungal treatments were reduced 79.5 and 84.5 percent, respectively. The apparent lack of effect following the second application can be attributed to a low CFU count of the second inoculum. Concentration was reduced from 5.4×10^5 CFUs/ml to 0.67×10^5 CFUs/ml. The effective concentration rates, taking into account the dilution factor, were 245 and 30 CFUs/ml for the first and second application, respectively. Effective concentration rates as low as 30 CFUs/ml would not be expected to produce the desired results.

Preliminary results from pond studies would seem to indicate that seasonality plays a role in susceptibility of hydrilla to fungal attack by the pathogen *M. terrestris*. Inoculum applied to field plots of hydrilla in August was effective in reducing aboveground biomass whereas applications made in June and July had no effect (Figure 2). Early in the season,

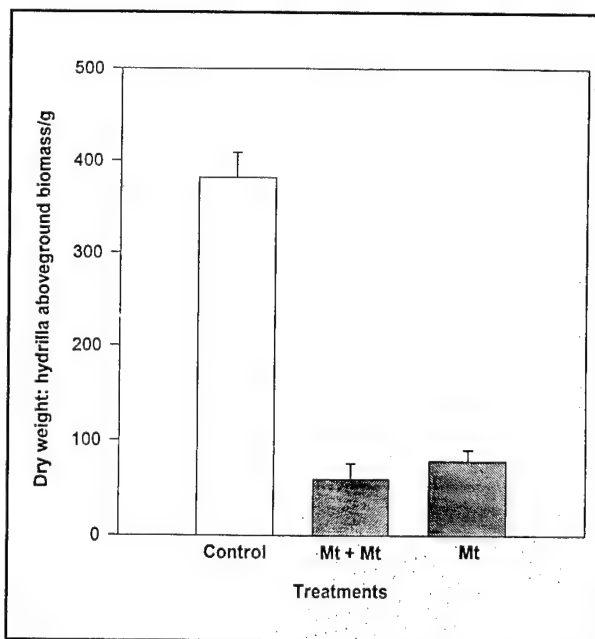


Figure 1. Mean dry weight in grams of aboveground biomass of Eurasian watermilfoil following one or two treatments with the fungus *Mycoleptodiscus terrestris*

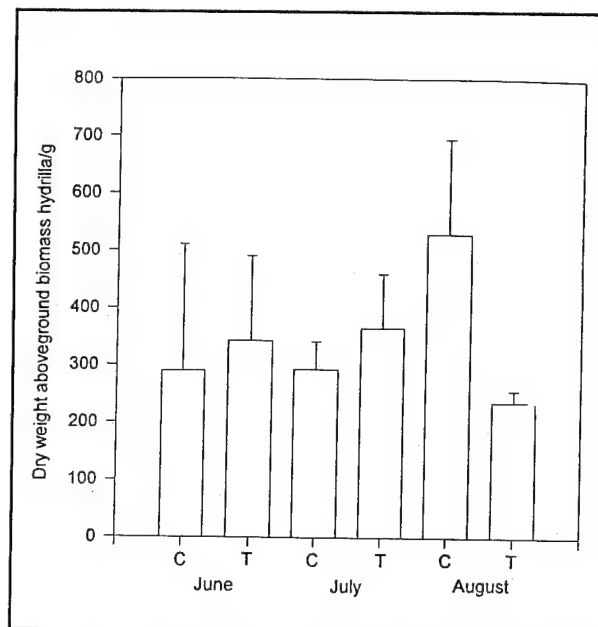


Figure 2. Mean dry weight in grams of aboveground biomass of hydrilla harvested 1 month post application from control and treated plots June, July, and August 1994

plant growth may exceed fungal growth to the extent that it severely limits the ability of the fungus to invade and ramify in-host tissue. Later in the growing season when new growth has slowed, the fungus may have time to become well established in plant tissues and induce chlorosis and fragmentation.

Additional studies must be forthcoming to confirm the preliminary results of seasonality and the effects of fungal applications. These studies, although time consuming, are extremely important in biocontrol management practices. Knowing that an agent is effective only at certain stages of plant growth must be well understood by applicators. In addition, knowing that a plant may be less susceptible at certain stages of its growth may allow an applicator to increase the dosage amount and bring about the desired results.

Myriophyllum spicatum

Based on laboratory results of efficacy testing on milfoil, isolates WES102292, WES19593, and WES59293 from Massachusetts, California, and Washington, respectively, were chosen for the field test at LAERF (Figure 3). A small portion of the inoculum prepared for the field application was reserved for testing the effects of the fungus on milfoil grown in 50-L aquaria. The CFU ratings of the inoculum were 5.3×10^5 , 3.3×10^5 , and 1.0×10^5 for isolates 102292, 19593, and 59293, respectively.

The aquarium experiment was harvested 2 weeks post inoculation because fungal impact on the milfoil was so severe that little plant tissue remained. All three isolates were effective in reducing aboveground biomass compared to the untreated controls even though there were some differences in the CFU ratings (Figure 4).

The same inoculum applied to field-grown milfoil was ineffective in reducing aboveground biomass even though the effective concentration following application was over five times that of the aquarium study (Figure 5). Poor field performance may be a factor of timing and site of application. Milfoil is more

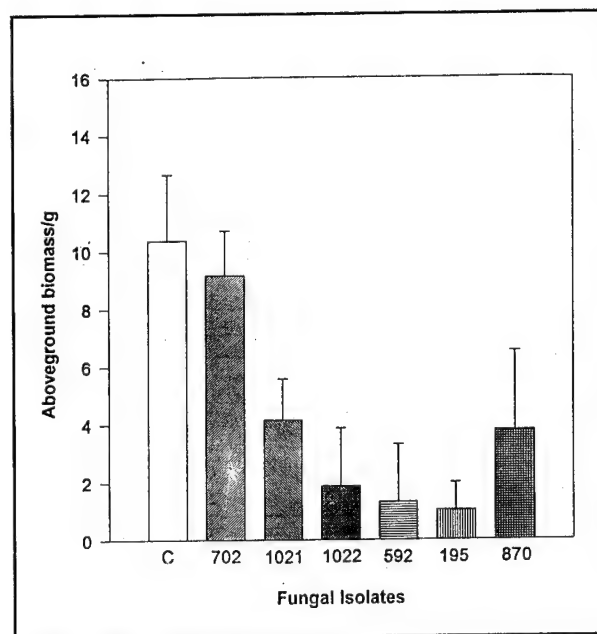


Figure 3. Mean dry weight in grams of aboveground biomass of Eurasian watermilfoil following inoculation with fungal isolates 70293, 102193, 102292, 59293, 19593, and 87093

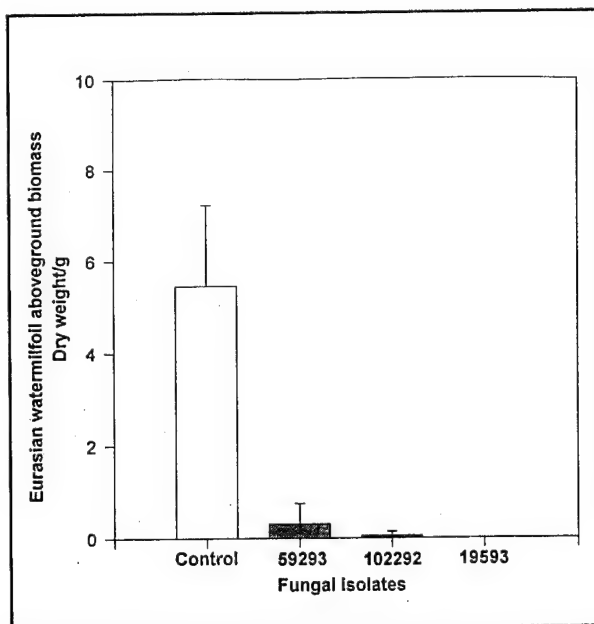


Figure 4. Mean dry weight in grams of aboveground biomass of Eurasian watermilfoil plots harvested 2 weeks post application with isolates 59293, 102292, and 19593

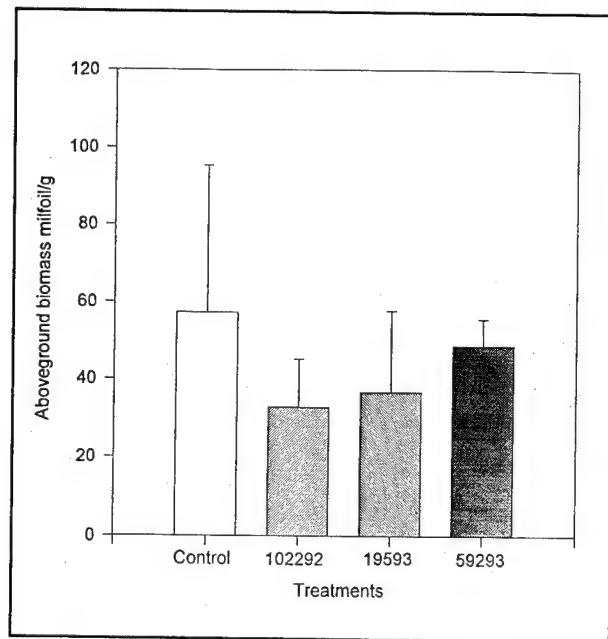


Figure 5. Mean dry weight in grams of above-ground biomass of Eurasian watermilfoil plots harvested 1 month post application with isolates 102292, 10593, and 59293

commonly found in cool temperate regions of the United States rather than warmer more southerly regions. Making a field application in Lewisville, TX, in August meant daily temperature maximums of 38 °C were not uncommon during the duration of the study. While the fungus will grow at high temperatures virulence is reduced at temperatures exceeding 30 °C. Milfoil under high temperature conditions was in a state of senescence and plant biomass between the plots was highly variable. Phenologically, inoculum might be more effective on milfoil if applied before the onset of senescence. In Texas, the timing of fungal applications should be early in the growing season, April or May, to avoid the temperature extremes present in milfoil ponds in mid to late summer.

Examination of the milfoil plants revealed an incrustation commonly referred to as marl surrounding the leaves and stems. Much of the incrustation was derived from epiphytic algal growth. For successful plant invasion to occur, direct contact must be made between the pathogen and the host tissues. The incrustation could provide a physical barrier which is impenetrable to fungal invasion or the sur-

face layer could have been inhibitory to fungal growth and development.

Foreign exploration/Asia

Foreign explorations for pathogens of Eurasian watermilfoil and hydrilla were initiated during 1994 in China. Cooperators and laboratory facilities were made available at the Sino-American Biological Control Lab in Beijing. During the initial visit, sites in and around Beijing were surveyed for populations of watermilfoil and hydrilla. Following field examination, plants showing any disease symptoms were collected and returned to the laboratory for microscopic examination and pathogen isolations. A total of 87 isolates were obtained from hydrilla, milfoil, and water. Tentatively identified were potential pathogenic species from the genera *Colletotrichum*, *Pythium*, *Sclerotium*, *Acremonium*, *Mycocleptodiscus*, and *Cylindrocarpon*. All isolates were transferred into axenic culture and sent to the pathogen quarantine facility at Fort Dietrich, MD, for accession into their collection.

Preliminary work on the collection of isolates from China will start at the pathogen quarantine facility. Identification of the isolates will be completed and those known to be pathogenic will be tested on the target plant and a range of nontarget plants. Only after an isolate is deemed safe will it be released from quarantine for additional research at the biocontrol laboratory at WES or in the field.

Future Work

Developmental work will continue on formulation designs for use in aquatic systems. Carriers with different adhering properties will be tested in the laboratory until a design is found which successfully meets the criteria for contacting and sticking to the plant. Work will also continue on fungal fermentation and preservation. Ideally the fermented fungal product must be produced in a cost-effective manner and still maintain its vigor, viability, and virulence. Following fermentation, techniques must be developed for incorporating

the fermented product into the carrier without losing any of those characteristics, a particularly difficult task when dealing with a living organism.

Field and laboratory testing of pathogens on hydrilla and milfoil will focus on plant phenology/pathogen effect, plant disease/dosage responses, and result discrepancies between laboratory/field testing. Laboratory work will also continue on screening new isolates acquired during 1994, culturing techniques, and host-specificity testing.

Isolates collected in China in 1994 and sent to Fort Dietrich will be processed at the quarantine facility. The work will entail isolate identification, screening of potential pathogens on milfoil and hydrilla, and host-specificity testing.

Survey work for biocontrol agents will continue in China on an expanded scale. We can now emphasize the team approach to the research efforts. The basis for this idea has come from Australia in their highly successful foreign exploration program for biocontrol

agents whereby they combine the expertise of a plant pathologist, an entomologist, and a plant ecologist. Because each of these scientists surveys or observes a plant community with a different focus the combined team effort is of great value for discovering natural controlling mechanisms of plants in their native ranges. Aquatic plant insect biocontrol researchers have been conducting overseas surveys for over 30 years. Since U.S. restrictions have eased on importation of exotic pathogens, and overseas exploration for pathogens of hydrilla and milfoil finally become a reality in 1994, we now have the opportunity to use the team approach in our efforts to acquire biocontrol agents. It is hoped that this approach will pay increasing dividends over the coming years.

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European Surveys for Pathogens of Eurasian Watermilfoil

by

Jo L. Harvey,¹ Duncan R. Varley,¹ and Harry C. Evans¹

Introduction

Myriophyllum spicatum, a member of the Haloragaceae family, is a submerged aquatic plant of Eurasian origin which has become a major weed in North American waterways. *M. spicatum* is widely distributed throughout the United Kingdom with records from Cornwall through to the Outer Hebrides, and occurs in most European countries from Scandinavia in the north to Sicily in the south (Kew Herbarium Records). It also occurs across Asia and in most of East Africa (Harley and Forno 1990). Eurasian watermilfoil grows in a wide range of environmental conditions, in both brackish and fresh water, from still water to fast-flowing streams and rivers, especially in lime-rich areas.

Other species that share its habitat include *Ceratophyllum* sp., *Myriophyllum alterniflorum*, and *M. proserpinacoides*.

M. spicatum has been a problem in the United States since the 1930's (Harley and Forno 1990). In the 1950's and 1960's it became a major ecological and economical weed in larger bodies of water, with reports from the Potomac River, Chesapeake Bay, and the Tennessee Valley Authority reservoirs (Bates, Hall, and Stanley 1972; Reed 1977). As an ecological problem, *M. spicatum* can greatly reduce the number of naturally occurring species. In Lake George (New York State), *M. spicatum* cover increased from 2 percent in 1987 to 20 to 45 percent in 1989; over the same period the number of other plant species fell from 20 to 9 (Madsen et al. 1990).

Use of pathogens has long been regarded as a good potential method of biological con-

trol for *M. spicatum* (Freeman and Charudattan 1980). Work has been undertaken on isolating and assessing fungal pathogens within the United States. Endophytic fungi have been reported in the literature on *M. spicatum*, in both Europe and the United States (Sparrow 1974; Luther 1979) and appear to be very damaging.

This is a report of the first of two seasons' surveys for pathogens, which concentrated on Southern England, Wales, Ireland, and Northern Europe. The second year, commencing 1995, will cover Northern England, Scotland, and Southern Europe.

The surveys and research work undertaken on this project were made possible through the sponsorship of the U.S. Government through its European Research Office of the U.S. Army.

Materials and Methods

Field surveys

The locations of potential sites of *M. spicatum* in the United Kingdom and the Republic of Ireland were obtained primarily from the Institute of Terrestrial Ecology database of aquatic flora. Additional information was obtained at a regional level from the National Rivers Authority, and on a local level from information centers and wardens of national parks.

Sites were selected in order to maximize the number of independent, environmentally diverse locations, rather than carrying out detailed surveys of any particular site. European sites were selected on identical criteria,

¹ International Institute of Biological Control, Silwood Park, Buckhurst Road, Ascot, Berkshire, U.K.

although with a greater emphasis on logistical requirements.

Sampling was carried out where possible by uprooting whole plants or clusters of plants and removing the surrounding soil by hand, thus minimizing mechanical damage. Where physical factors prevented this approach, a miniature grappling hook, or weed drag, was utilized, enabling rooted samples to be obtained in most cases. Where appropriate, waders and/or a rowing boat were employed.

Plant material was collected in reusable, sealable pots, and/or plastic ziplock bags, which were filled with water from the collection site. Soil material, where possible to obtain, was stored in plastic universal bottles, as were water samples used for measuring pH.

Laboratory studies

Isolations were made from plant material that had been washed for two to three hours, under running tap water. Sections of leaves, roots, and stems were plated out onto tap water agar (TWA) containing penicillin and streptomycin sulfate antibiotics. Isolations from soil were made on TWA containing antibiotics and Komada's medium (Komada 1975). Isolations from water were made using a range of baiting techniques and media selective for Oomycetes (Ribeiro 1978).

Subcultures were maintained at 20 °C, on potato carrot agar containing antibiotics. Blacklight on a 12-hr cycle was used to stimulate sporulation.

Provisional identifications were made at the International Institute of Biological Control (IIBC), and confirmed at the International Mycological Institute.

Cultures were stored on PCA at 10 °C until screening.

Results and Discussion

Species and genera

Table 1 shows the range of species and genera of fungi isolated and identified to date from *M. spicatum*, over the course of surveys.

Ecology

Myriophyllum spicatum, as expected, occurred in a wide range of environmental conditions.

There was, however, an apparent tendency for it to occur in more alkaline waters, although the lowest pH level recorded was 5.5, the range being from 5.5 to 9.8. It also appeared to grow preferentially in unshaded areas and this was apparently a critical factor delimiting its occurrence. Growth was observed in open waters or in areas of water without shading.

Eurasian watermilfoil was found to occur in still water: canals, e.g., Grand Canal, Ireland; drainage ditches, e.g., Chantry Point Ditch, Suffolk, U.K.; lakes, e.g., Lake Geneva, Switzerland; ponds, e.g., Roe Ponds, Derbyshire, U.K.; reservoirs, e.g., Feilinger See, Germany; in slow-moving rivers, e.g., River Frome, Dorset, U.K.; and fast-flowing streams, e.g., Cherrybrook, Dartmoor, U.K.

It occurred at depths from 0.05 m, Dockens Water, Hampshire, U.K., a fast-flowing stream, to 2.0 m, Slapton Ley, Devon, U.K. Depth was observed to be a limiting factor, clearly visible in individual lakes, e.g. Grasmere, Cumbria, U.K. Here, *M. spicatum* was seen growing on a steep gradient bank, a clear line visible where growth was no longer present below a depth of around 1 m.

Several different growth forms of *M. spicatum* were observed under field conditions. Variations in leaf thickness, internodal distance,

Table 1**A List of Genera and Species Isolated and Identified from European Surveys
(Sep 1993-Oct 1994)**

<i>Absidia cylindrospora</i>	<i>Fusarium acuminatum</i>	<i>Nectria discophora</i> (anamorph)
<i>Acremonium persicinum</i>	<i>F. crookwellense</i>	<i>Phoma complanata</i>
<i>Acremonium strictum</i>	<i>F. culmorum</i>	<i>P. eupyrena</i>
<i>Alternaria infectoria</i>	<i>F. equiseti</i>	<i>P. exigua</i>
<i>Ascochyta</i> sp.	<i>F. flocciferum</i>	<i>P. macrostroma</i>
<i>Aureobasidium</i> sp.	<i>F. graminearum</i>	<i>P. leveillei</i>
<i>Byssochlamys nivea</i>	<i>F. oxysporum</i>	<i>Plectosphaerella cucumerina</i>
<i>Botrytis cinerea</i>	<i>F. polyphialides</i>	<i>Pythium</i> sp.
<i>Chrysosporium</i> sp.	<i>F. poae</i>	<i>Pythium</i> sp. group T
<i>Colletotrichum</i> sp.	<i>F. sambucinum</i>	<i>P. aquatile</i>
<i>Coniothyrium sporulosum</i>	<i>F. sporotrichoides</i>	<i>P. periplocum</i>
<i>Corynascus sepedonium</i>	<i>Gliocladium roseum</i>	<i>P. scleroteichum</i>
<i>Cryptosporiopsis</i> sp.	<i>Gliomastrix murorum</i> var <i>felina</i>	<i>Stagonospora</i> sp.
<i>Cylindrocarpon destructans</i>	<i>Glomerella cingulata</i>	<i>Saprolegnia parasitica</i>
<i>Cylindrocarpon ianthothele</i>	<i>Microdochium tabacinum</i>	<i>Trichosporiella sporotrichoides</i>
<i>Cylindrocladium</i> sp.	<i>Microsphaeropsis</i> sp.	<i>Verticillium nigrescens</i>
<i>Embellisia</i> sp.	<i>Mycocentrospora acerina</i>	
<i>Emercelloopsis minima</i>	<i>Myrothecium cinctum</i>	

overall length, and plant spacing appeared to be dependent on local conditions, and are assumed to be adaptations to these conditions. The most notable variation in growth was found in Derwent Water, Cumbria, U.K., where *M. spicatum* appeared to be restricted to the bottom 0.2 m, in a depth of 0.7 m. In all cases, the samples taken reverted to a standard form of growth when grown in tanks under laboratory conditions; an ecotypic rather than a biotypic character.

It was not possible to determine whether factors such as light intensity, carbonate levels, dissolved carbon dioxide levels, nutrient composition, or temperature were limiting the occurrence of *M. spicatum* in this study.

Conclusions

A range of potential plant pathogenic species and genera have been isolated and identified over the course of the surveys. These include several genera which have previously shown some response in the United States: *Acremonium* sp. (Andrews, Hecht, and Bashirian 1982); *Colletotrichum* sp. (Smith et al. 1989); and *Fusarium* sp. (Andrews and Hecht 1981). Pathogens isolated from the plant at the center of origin should be better adapted, more specific, and, therefore, have a potentially greater effect than supposedly less specific or opportunistic U.S. pathogens.

During the surveys, *M. spicatum* was never observed to be invasive or a weed problem, except in very heavily disturbed areas, and this would suggest that natural control is in operation.

The relatively recent arrival in outbreak form of the North American *M. heterophyllum* in areas of Central Western Europe, as well as the absence of *Mycroplectodiscus terrestris* from the list of fungi isolated from *M. spicatum* in Europe, would suggest that there are two sets of mycoflora, one from the center of origin and one in the United States.

It can be concluded, therefore, that there is great potential for biological control, and this has been reflected in early success with a species of *Acremonium* during the initial pathogenicity screening of isolates collected.

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Current Status on the Use of Insect Biocontrol Agents for the Management of Hydrilla

by

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Introduction

The operational use of insect biocontrol technology for the management of hydrilla began in Florida during 1987 with the release of *Bagous affinis*, the hydrilla tuber-feeding weevil. Since that initial release, three additional agents for the management of hydrilla have been introduced in the Southeast as well as various parts of California. The hydrilla biocontrol program has seen tremendous growth with over 4 million individuals of the four agents introduced using a combination of laboratory/greenhouse mass-rearing techniques as well as field collection procedures (Center 1989; Center and Dray 1990; Center, Dray, and Durden 1991).

This research has been summarized in much detail in the last several volumes of the Proceedings of the Annual Meeting, Aquatic Plant Control Research Program Review (Grodowitz et al. 1994; Grodowitz et al. 1993; Center 1992; Grodowitz and Snoddy 1992). These summaries have included information on the agent's life history, past and present release efforts, as well as establishment status. This present paper is an effort to summarize the work conducted in 1994 concerning the release and establishment of insect biocontrol agents of hydrilla in the United States. It is divided into separate sections with each section summarizing the release and establishment work for each agent.

The Leaf-Mining Flies *Hydrellia pakistanae* and *Hydrellia balciunasi*

Most of the work accomplished for the introduced *Hydrellia* during 1994 has concentrated on monitoring population levels and their associated damage at various U.S. sites. The monitoring has included both qualitative and quantitative efforts. The majority of the quantitative monitoring was accomplished at sites in Alabama and Texas. In addition, limited field releases were made at several locations in Alabama and Texas.

Since the first release of *H. pakistanae* in 1987 higher numbers of individuals have been released with each successive year up to 1993 (Figure 1). The significant increase in numbers released in 1993 relative to preceding years is due partly to the methods used for obtaining insect release material. In 1993, the largest releases were made by harvesting *H. pakistanae* infested plant material collected from ponds in Muscle Shoals, AL, and transporting it to sites in Georgia (i.e., Lake Seminole) and Louisiana (i.e., Lake Boeuf) (Grodowitz et al. 1994). Total number of immature *H. pakistanae* released using this pond-harvesting method was estimated to be over 2 million individuals.

This is contrasted to releases made in 1994. For 1994, releases were made primarily from

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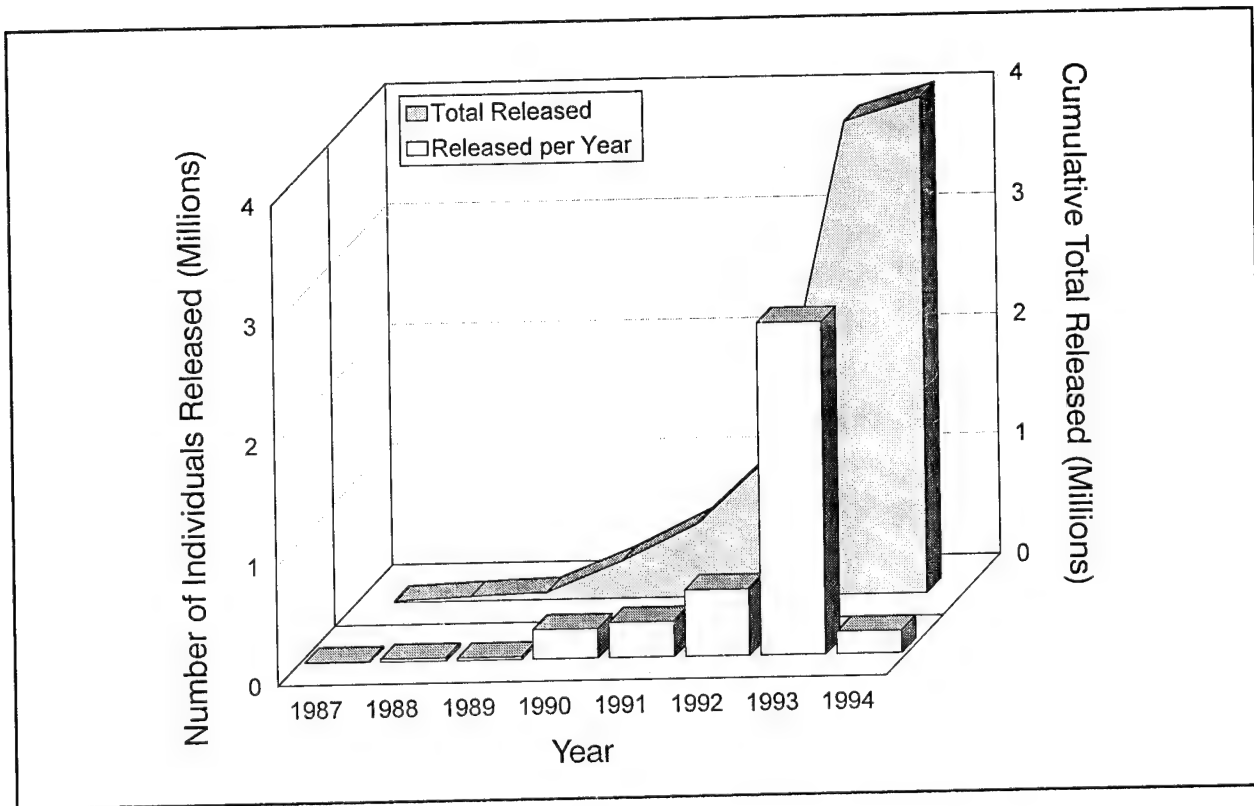


Figure 1. Total number of *H. pakistanae* individuals released per year and cumulative total released since 1987

insects obtained from mass-rearing facilities housed at WES, Vicksburg, MS, and the TVA Reservation in Muscle Shoals, AL. In addition to laboratory/greenhouse mass-rearing efforts, first generation larvae used for the releases were obtained from adults collected from field sites in Florida and Alabama.

In 1994, releases of *H. pakistanae* were made at Coletto Creek Reservoir and Choke Canyon Reservoir both located near Corpus Christi, TX. In addition, releases were made at the Sublette Ferry site on Lake Guntersville, AL (Table 1). These releases resulted in establishment of *H. pakistanae* at sites on

Table 1
Releases of the Introduced *Hydrellia* sp. Made During 1994

Species	Number of Releases	State	Site	Release Method	Number Released
<i>H. pakistanae</i>	1	TX	Coletto Creek Reservoir	Field Collection	60,000
<i>H. pakistanae</i>	1	TX	Choke Canyon Reservoir	Field Collection	36,000
<i>H. pakistanae</i>	19	AL	Sublette Ferry	Mass-Rearing	92,518
				TOTAL	188,518
<i>H. balciunasi</i>	2	TX	Choke Canyon	Mass-Rearing	9,740
<i>H. balciunasi</i>	1	TX	Huntsville State Park	Mass-Rearing	2,048
				TOTAL	11,788

Coletto Creek Reservoir and increased populations of *H. pakistanae* at Sublette Ferry.

Releases of *H. balciunasi*, a species closely related to *H. pakistanae*, were limited to only two sites and less than 12,000 individuals. In addition, over 15,000 additional individuals were used for experimental purposes by USDA/ARS personnel, Fort Lauderdale, FL, and by researchers at WES.

Populations of *H. pakistanae* have continued to spread to areas outside their original release. In Florida, collections of leaf-mining flies from hydrilla infestations from the southern tip of the peninsula north to Gainesville usually yield at least some individuals of *H. pakistanae*. In some areas in southern Florida, populations have been at levels high enough to effect visible damage to the hydrilla. This indicates that this species is firmly and permanently established throughout the southern portion of the state. Small populations of *H. pakistanae* can also be found as far west in Florida as Lake Seminole. Collections from this lake, which is located on the Georgia-Florida border, demonstrate the presence of small but permanently established populations at several locations.

H. pakistanae can also be found as far north as Muscle Shoals and Lake Guntersville, AL. Populations have occurred in ponds on the TVA Reservation in Muscle Shoals for the last 3 years while the population on Lake Guntersville has been established since 1993. Some of the highest documented population levels and associated damage for *H. pakistanae* have been recorded at the Muscle Shoals pond facility (Grodowitz et al. 1994).

H. pakistanae can also be collected as far west as Coletto Creek Reservoir. This reservoir is approximately 100 miles northeast of Corpus Christi. The newly established population resulted from introducing larvae reared from field-collected adults gathered from Florida sites. A single release was made on Coletto Creek Reservoir during the spring of 1994 and *H. pakistanae* individuals have been collected from this site during recent sampling efforts (i.e., December 1994).

H. pakistanae has been found to reach high and damaging population levels in relatively short time periods (Figure 2). Even after initial releases of only about 50,000 flies, high and damaging populations have been found at many of the ponds at the TVA Reservation planted with hydrilla during 1992. High numbers of immatures and associated damage continued for the 1993 growing season. Additional releases were made in several adjacent ponds during the summer of 1993. Immatures released during 1993 numbered about 30,000 individuals.

Population numbers and associated leaf damage for the last three growing seasons for *H. pakistanae* on ponds 9, 19, 20, and 21 (i.e., the original hydrilla ponds) in the TVA pond facility in Muscle Shoals have shown significant ($p < 0.0001$) decreases over the last 3 years (Figures 3 and 4). The growing season is defined as July, August, and September. Reasons for limited population increases during 1994 and to some extent in 1993 are unknown but may be related to the minimal numbers of immatures released during 1994 coupled with periods of harsh environmental conditions. There were only small augmentative releases ($< 2,000$) made on the TVA pond facility during 1994 as compared to 50,000 during 1992 and 30,000 for 1993. In addition, the 1994 growing season followed a relatively harsh winter with several periods of ice formation occurring on the ponds and water temperatures reaching 4 °C during extended periods in January and February.

While large and rapid population increases have been shown for *H. pakistanae* only limited increases have occurred for *H. balciunasi* populations (Figure 5). Since the first releases of *H. balciunasi*, this species has proven to be exceptionally hard to establish even with repeated attempts in Florida and Texas (Grodowitz et al. 1994; Grodowitz et al. 1993; Center 1992; Grodowitz and Snoddy 1992). To date, only one site, i.e., Sheldon Reservoir, has had permanently established populations for any extended periods (Figure 5). As of this summer, *H. balciunasi* individuals have commonly been collected from another Texas release site, i.e., Huntsville State Park. While populations at Huntsville State Park appear

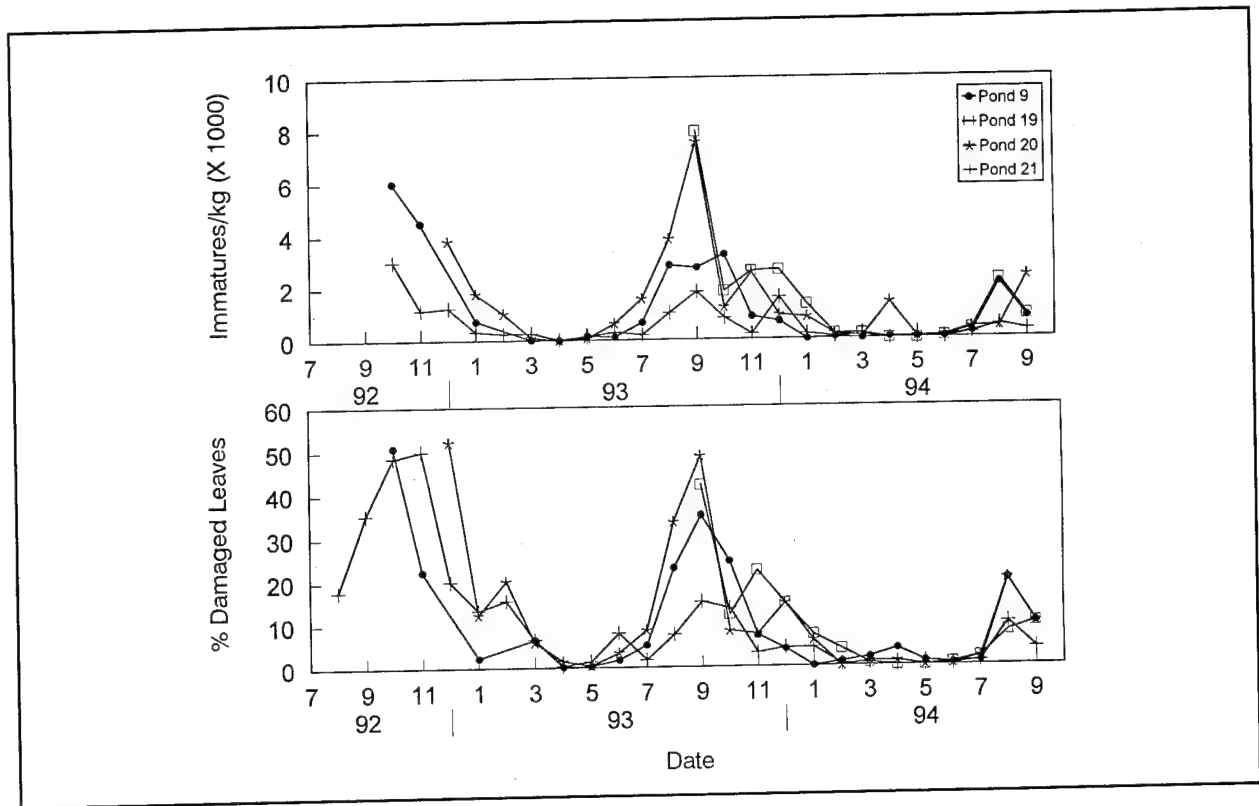


Figure 2. Total number of associated immature *H. pakistanae* and percent leaf damage for ponds on the TVA Reservation, Muscle Shoals, AL

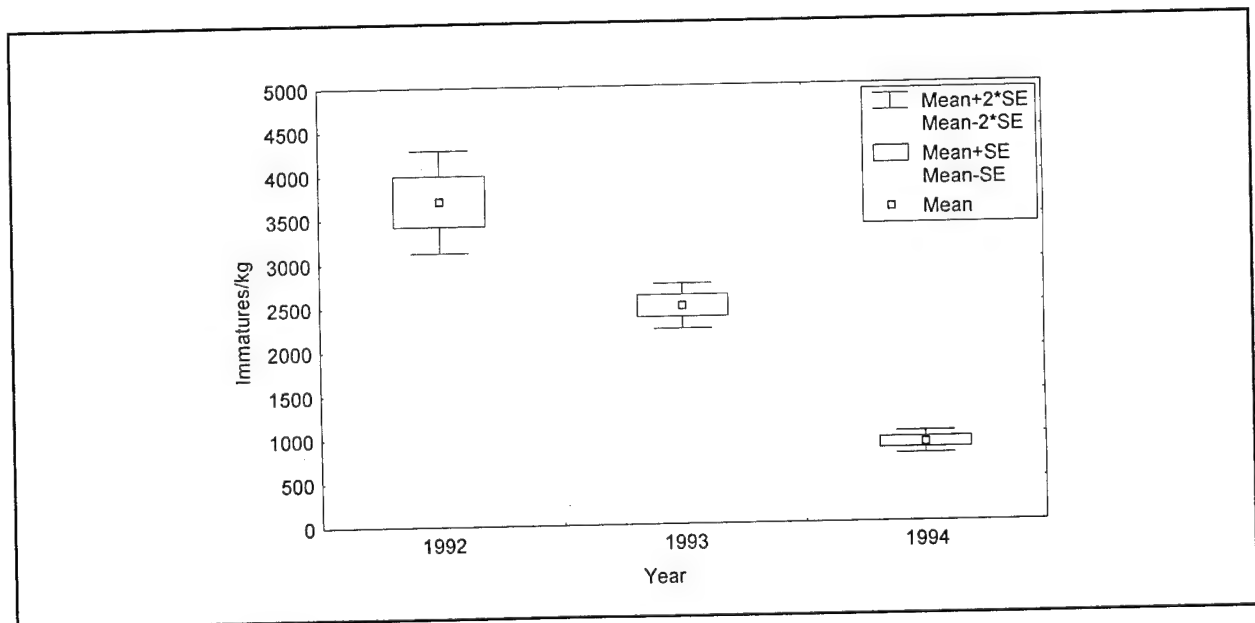


Figure 3. Average number of immatures from the active growing season (i.e., July, August, and September) for ponds on the TVA Reservation, Muscle Shoals, AL

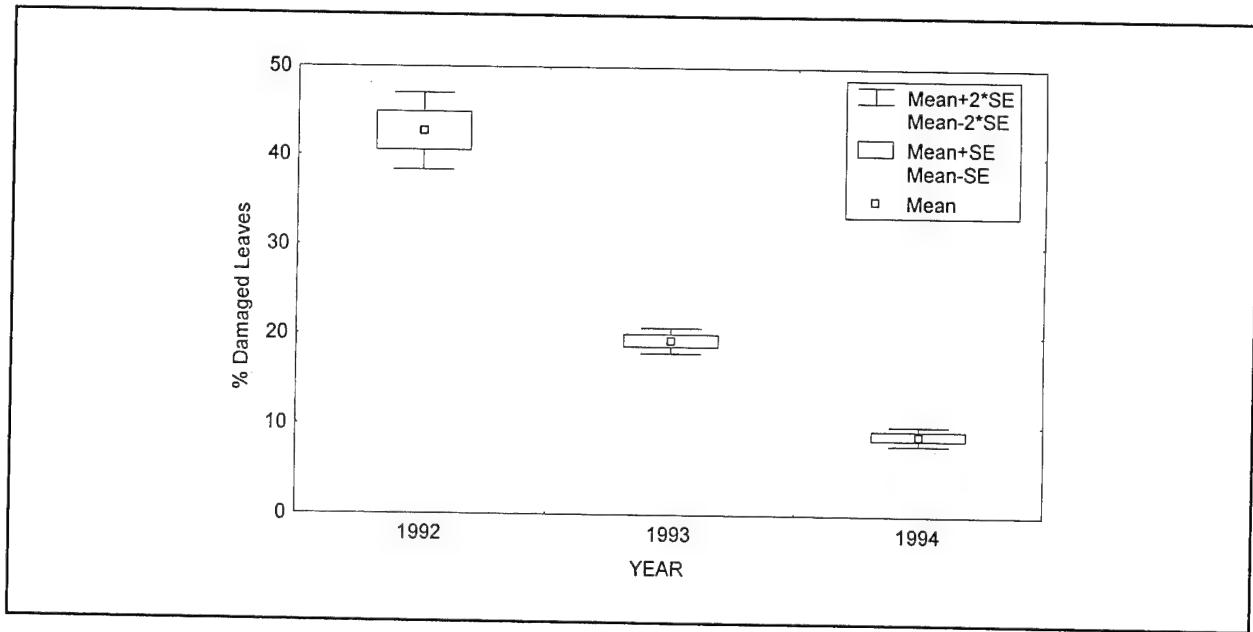


Figure 4. Average percentage leaf damage for *H. pakistanae* infested hydrilla from the active growing season (i.e., July, August, and September) for ponds on the TVA Reservation, Muscle Shoals, AL

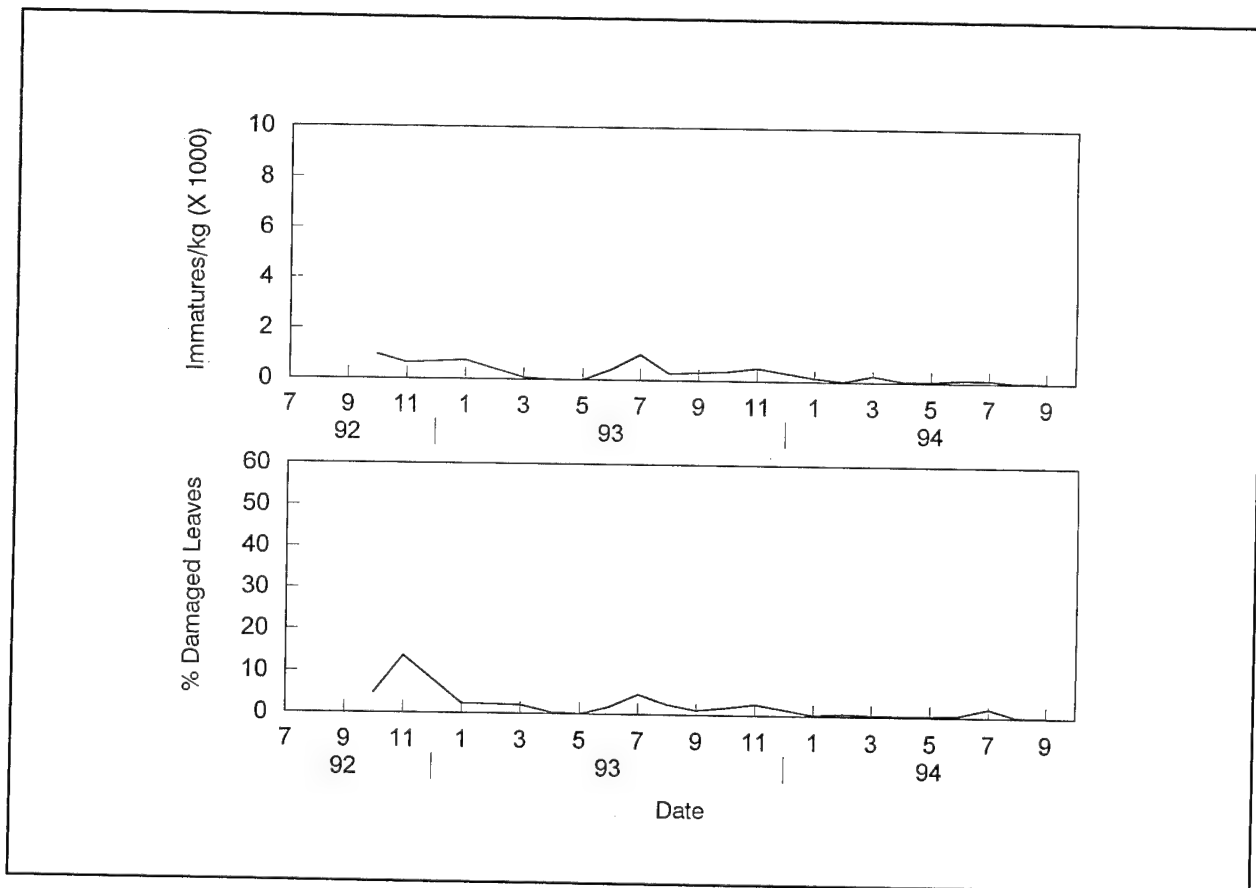


Figure 5. Total number of immature *H. balciunasi* and associated percent leaf damage for Sheldon reservoir near Houston, TX

small, based on only limited number of individuals collected at any given time, the consistency of collections during summer and fall of 1994 indicates that *H. balciunasi* is permanently established at this site. Reasons for such difficulties in establishing this species are unknown but may be due to stricter requirements for this species in terms of hydrilla leaf hardness, preferring, instead, softer leaf biotypes. More research is needed to explain such differences. Preliminary attempts at explaining the lack of establishment can be found in another paper included in this proceedings (Wheeler and Center 1995).

The Stem-Feeding Weevil *Bagous hydrillae*

The first release of the stem-feeding weevil, *Bagous hydrillae*, was made in Florida during 1991. Since that initial release close to 300,000 individuals have been introduced in four states at 14 different sites. A variety of different methods have been attempted for successfully establishing this species (Grodowitz et al. 1994).

However, from the onset of release efforts in Florida this species has proven to be extremely difficult to establish (Grodowitz et al. 1994). It is believed that this difficulty is due to strict environmental conditions required by this species for successful completion of development. While only limited information is available on *B. hydrillae* life history, evidence from overseas and quarantine research, as well as initial release attempts in the United States, indicate that relatively dry soil conditions and large quantities of hydrilla stranded on the shoreline are needed for proper development and, ultimately, pupation.

Unfortunately, such conditions are typically not found in most of the southeastern United States. However, during release and establishment attempts for the two species of leaf-mining flies, a site in Texas was found with these desired hydrilla characteristics. Choke Canyon Reservoir is about 80 miles northwest of Corpus Christi at the confluence of the Frio, Nueces, and Atascosa Rivers. Downstream from Choke Canyon Reservoir is

Lake Corpus Christi, a large reservoir which serves as a source of water for local municipalities and industries. Over the last 1.5 years this area has had significant decreases in local rainfall which is impacting water uses for these two water bodies by the dropping of water levels in both reservoirs. In addition, Lake Corpus Christi must maintain water levels of 88 ft to allow for water uptake for various uses. To this end, water is pumped from Choke Canyon Reservoir to Lake Corpus Christi in order to maintain the required 88-ft level in Lake Corpus Christi. This has resulted in large areas of hydrilla to become de-watered and thereby form large and extensive strandlines of drying hydrilla along the new shoreline of Choke Canyon (Figure 6).

Beginning in 1993 and continuing through fall of 1994 close to 40,000 *B. hydrillae* adults have been released at four different locations on Choke Canyon Reservoir. The most recent release was during the latter half of September 1994. Since that last release, *B. hydrillae* adults have routinely been collected from areas near the original introductions. Adults have been collected by hand as well as extracted from collected plant material using Berlese funnels. It appears that this species is tentatively established at Choke Canyon. This area will be sampled monthly to determine the overwintering capacity of the species in southern Texas. The population will be considered permanently established if living adults can be collected after extended periods with no additional releases. The unique characteristics of this site have apparently allowed this species to become established.

The Tuber-Feeding Weevil *Bagous affinis*

The first species released for the management of hydrilla was the tuber-feeding weevil, *Bagous affinis*. It was first released in Florida during the spring of 1987. Releases continued in Florida until 1988 when further release attempts were discontinued. Releases were discontinued because of highly specific environmental conditions required by this species for growth and development. *Bagous affinis*



Figure 6. Photograph of Choke Canyon Reservoir, TX, taken during 1994, showing stranded hydrilla drying along the shoreline

requires extensive drawdown conditions which expose the sediment surface and allow immatures access to buried tubers, the primary food source for the immatures. Such conditions are rarely found in the Southeast. Beginning in 1992, *B. affinis* was released at sites in northern California (Godfrey et al. 1994). Water levels at many sites in California can be manipulated to a large extent thereby allowing for the formation of site characteristics conducive for *B. affinis* growth and development.

However, Choke Canyon Reservoir, with its extensive natural and man-made de-watering over the last 1.5 years, appeared ideal for the release of *B. affinis*. The site had extensive areas of exposed sediment with tuber densities averaging at least 90 tubers per m². During September 1994 about 200 *B. affinis* were released into a small cage. The site appeared to be in excellent shape as indicated by numerous

tubers present in the first few centimeters of the sediment. Several tubers were observed to be germinating. During December 1994, the cage was removed and the entire sediment to a depth of about 10 cm removed and searched for the presence of *B. affinis* as well as insect damaged tubers. While no signs of the original individuals were found or their associated damage we are still hopeful that the specific characteristics of this site (i.e., large areas of exposed sediment) will allow the establishment of this species where other less suitable sites have not allowed establishment. To further strengthen our chances for establishment success we are obtaining fresh stock of *B. affinis* from India for use in releases to be made during late spring and summer 1995 on Choke Canyon Reservoir. Releasing newly collected material from the insect's home range typically increases chances for establishment.

Summary

The release and establishment program for insect biocontrol agents of hydrilla has grown considerably since the first agents were released in 1987. During 1994, we observed significant range extensions throughout Florida, north to Muscle Shoals, AL, and west to Col-eto Creek Reservoirs, TX, for *H. pakistanae*. During 1993 and 1994, high populations of *H. pakistanae* were correlated with significant impact to hydrilla at sites in Muscle Shoals and at various locations in southern Florida.

It is discouraging, however, that only limited establishment has been observed for *H. balciunasi*. More foundational research is underway to determine what environmental factors are important in limiting population establishment, expansion, and growth at specific sites for this species. Such information may allow for *H. balciunasi* establishment at other sites in the future.

Bagous hydrillae appears to be established on Choke Canyon Reservoir but verification is needed and is expected during late spring or summer 1995. This same site also appears to have excellent characteristics for future establishment of the tuber-feeding weevil, *B. affinis*.

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Hydrilla Leaf Toughness Affects Larval Growth and Development of the Biocontrol Agent *Hydrellia pakistanae*

Gregory S. Wheeler¹ and Ted D. Center²

Introduction

Plant quality can influence host acceptability and herbivore growth and development in both terrestrial (Rosenthal and Berenbaum 1991, 1992; Stamp and Casey 1993) and aquatic environments (Center and Wright 1991; Taylor 1984). Herbivore survival, growth, and development can be influenced by plant nutrients, defensive compounds, and/or tissue toughness. Among these factors, possibly the least studied is tissue toughness which may have a dramatic effect on herbivores by interfering with ingestion, digestion, or physically restricting the feeding activities of external or internal plant feeders (Feeny 1970; Raupp 1985; Lindroth, Kinney, and Platz 1993).

Preliminary surveys of hydrilla revealed that leaf toughness varies considerably during different seasons, at different locations in Florida, and within the same location. For example, leaf toughness of hydrilla collected at different locations in Florida ranged from about 45 to 200 g/mm² (the force required for leaf penetration by a penetrometer (e.g., Lindroth, Kinney, and Platz 1993). The larvae of the biocontrol agent *Hydrellia pakistanae* feed and tunnel inside hydrilla leaves and, thus, the tougher hydrilla leaves may reduce their growth and development. The goal of this study was to determine the effect of different hydrilla leaf toughnesses on *H. pakistanae* larval survival, growth, and development.

Results

Leaf toughness

Two distinct hydrilla leaf toughnesses were found at the Miami Canal in Broward Co., FL, during August 1994. The leaves of the tough hydrilla were consistently tougher than those of the soft hydrilla at all whorl locations starting with the apical tip down to 15 cm. Furthermore, leaf toughness increased for both tough and soft hydrilla from the apical tip (whorl 0) toward the stem base (whorl 15).

H. pakistanae larval survival, growth, and development

When first instar *H. pakistanae* were fed either the tough or soft hydrilla and reared to the adult stage their performance was reduced. Percent mortality of the larvae reared on the tough hydrilla (60 percent) was greater than for those reared on the soft hydrilla (<10 percent). For larvae reared on either hydrilla tissue type, most mortality occurred during the first instar; however, no larvae died during the third instar. Furthermore, the larvae fed the tough hydrilla took longer to develop to the pupal and, hence, adult stages compared with those fed the soft hydrilla. Adult female biomass was also reduced for larvae fed the tough hydrilla. Additionally, females in general, regardless of diet, had greater biomass than males.

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***H. pakistanae* larval and pupal location**

Hydrilla type also influenced the location of fly larval feeding and pupation. Each larval instar occurred most frequently in the apical leaves (whorl 0) in the tough hydrilla, whereas they occurred most frequently in whorl 5 in the soft hydrilla. Pupation similarly occurred most frequently in whorl 0 in the tough, compared with whorl 5 in the soft hydrilla. The regularity of the whorl occupied by larvae and pupae, fed either hydrilla type, did not change during the course of the experiment.

Discussion

Among the factors that influence the success of the establishment of biocontrol agents (e.g., climate, natural enemies), the nutritional status of the host plant frequently plays a key role (Myers 1987; Julien 1992). Several examples may be cited from the classical successes in biocontrol of weeds that support this contention. Among these are the *Cactoblastis* moth on prickly pear cactus (Dodd 1940) and the *Cyrtobagous* weevil on the *Salvinia* aquatic fern (Thomas and Room 1986), both of which were successfully established only after the initial applications of nitrogen fertilizer. We are aware of only one study that investigated the influence of tissue toughness on the success and establishment of a biocontrol agent. Waterhyacinth petiole hardness was found to reduce tissue penetration by the larvae of *Sameodes albiguttalis* (Wright and Bourne 1986). Our results indicate that hydrilla leaf toughness has a dramatic effect on the survival, growth, and development of the biocontrol agent *H. pakistanae*. Larval mortality of first instars and pupal and adult developmental times increased when larvae were fed tough hydrilla leaves. The biomass of adult females decreased when larvae were fed tough hydrilla leaves. Furthermore, when the first instar larvae were fed only tough leaves they appeared to select the softer leaves at the terminus of the hydrilla plant for feeding.

The impact of this biocontrol agent on hydrilla plants is influenced by a number of

plant nutritional factors, among them leaf toughness. Additional studies are needed to evaluate the effect of other nutritional factors such as foliar nutrients and defensive chemistry on the performance of these biocontrol agents. Our results indicate that when *H. pakistanae* larvae were offered only the coarse hydrilla type their performance decreased. However, in the field where considerable variation in leaf toughness occurs, the selection behavior exhibited by these larvae may enable them to exploit the softest available tissues, thereby mitigating the negative impact of hydrilla leaf toughness.

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Temperature Study of *Hydrellia pakistanae*

by
Ramona H. Warren¹

Introduction

Background information

Hydrilla (*Hydrilla verticillata* (L.f.) L. C.) is a submersed aquatic plant with a high reproductive potential and wide ecological tolerance. It reproduces both vegetatively and by seeds (Baloch and Ullah 1973). The plant is found throughout the southern United States and along the East Coast as far north as Delaware. Populations of hydrilla are also found in California (Cofrancesco 1991).

This nuisance aquatic plant causes problems by interfering with waterflow and recreation. It also impedes navigation by clogging waterways.

Mechanical and chemical applications are used as control alternatives and appear to be effective. In areas where there are large plant infestations, mechanical harvesters are not as effective or efficient as other control methods. Chemical applications were effective, but primarily as a maintenance measure (Hamilton 1991).

The development of biocontrol technology began in 1959 when the Corps of Engineers and the USDA entered into a cooperative study to manage exotic aquatic plants (Cofrancesco 1991). Biocontrol methods such as insects seem to present the most perplexing short-term problems, but also seem to hold the most hope for long-term solutions (Hamilton 1991). *Hydrellia pakistanae*, an ephydrid fly for hydrilla from Pakistan was the second insect released in the United States in Florida. The four life stages are eggs, larvae, pupae, and adults (Figure 1). The larvae mine the leaves

of hydrilla and larva is the only life stage that impacts the plant (Buckingham and Okrah, in preparation).

Objective

The impact of temperature on developmental and survival rates of *H. pakistanae* is important because insect biocontrol agents for *hydrilla* have been released and evaluated primarily in Florida, and now that researchers have begun to release flies as far north as Guntersville, AL, it is important to know if the insects can establish and survive over winter in a large lake system such as Guntersville Reservoir. The studies reported in this paper will provide background information necessary for the selection of release techniques that are optimal for establishment of the insects.

Methods and Materials

The experiment was initiated by obtaining insect eggs from the WES and the TVA colony of *H. pakistanae*. *Hydrilla* sprigs were obtained from the WES greenhouse facilities for use in this experiment. Several different techniques were tested for developmental and cold temperature studies so that a protocol for the studies could be developed.

The objectives of the developmental studies were to (1) examine rate of development at different temperatures, (2) determine leaf damage, and (3) determine percent survival. The objective of the cold temperature study was to determine percent survival at different life stages when exposed to cold temperature for a certain period of time.

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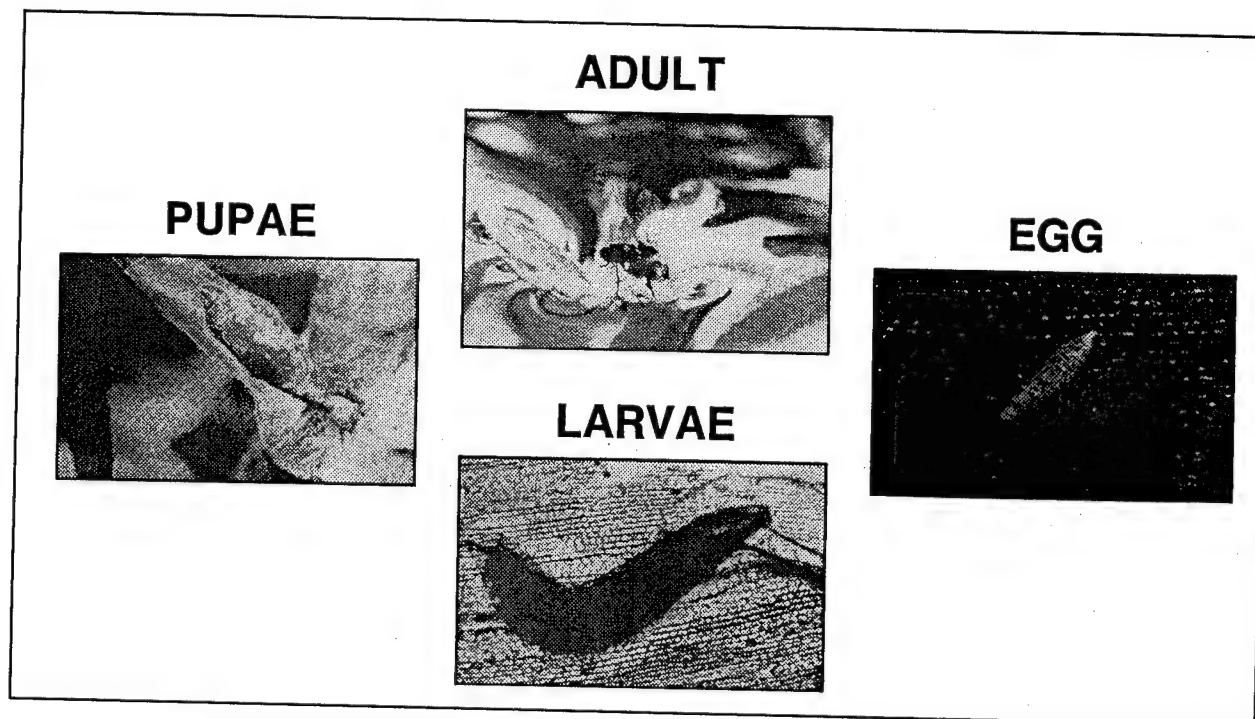


Figure 1. Life stages of *H. pakistanae*

Developmental studies

Nine sprigs of *hydrilla* were placed in a petri dish and put into the colony box for egg deposit over a 24-hr time period. The eggs were then retrieved and placed in petri dishes that contained a sheet of black filter paper and 20 individual *hydrilla* leaves with one egg on each leaf (the black filter paper allowed us to see the egg easily). These petri dishes were then placed in an environmental chamber at the desired test temperature ranging from 13 to 35 °C with a 14-hr photophase and were checked daily for the first instar larvae.

One 15-cm sprig of *hydrilla* with 15 to 20 whorls of four or five leaves was placed in each of forty 60-ml test tubes containing deionized water. One first instar larva was placed in each test tube. These tubes were covered with nylon organdy and held in place with a rubber band. (Note: The tubes were placed in an environmental chamber at the same time as the five petri dishes so that the sprigs could adjust to the desired temperature.) The test tubes were examined daily, using a dissecting microscope, for (1) development of life stage and (2) location and timing for the duration of each temperatures test.

The data were analyzed using a statistical program.

Cold temperature study

The experiment was initiated by using sixty 40-qt mason jars filled with deionized water and 45 g of *hydrilla*. Thirty insects of the different species were placed in the jars according to species (*H. pakistanae*; China strain of *H. pakistanae*, and *H. balciunasi*). Three environmental chambers were used in this experiment: A holding chamber at 27 °C, a cold chamber at 6 °C, and a ramping chamber from 6 to 27 °C over a 2- to 3-day period. The ramping chamber would allow the larvae time to adapt to the warmer temperature after being moved from the cold chamber, similar to that of a natural environment. All chambers remained at the temperature constant. The holding chamber was where the control was placed from the very beginning of the experiment which was 0 week. The rest of the jars were placed in the cold chamber at 6 °C for exposure times of 1, 2, 4, and 8 weeks. After the exposure times were completed the jars were taken from the cold chamber and placed in the ramping chamber for 2 to 3 days. They were then placed in the holding chamber where they

remained until they developed into adults. The jars were checked daily for emergence of adults, and percent survival was recorded.

Results and Discussion

Developmental studies

This research showed that shorter developmental rates were associated with higher temperature (Figure 2). There was a good trend between all temperatures tested, but 30 and 33 °C showed the greatest trend for all the temperatures tested. At 35 °C development time began to rise again, which could possibly mean this is the threshold limit. When we looked at the various life stages we found basically the same trends as the overall development. At the egg stage (Figure 3), the greatest trend was at 30 °C. At first instar the best trend was at 33 °C (Figure 4). For second and third instar the greatest trends were at 30 and 33 °C (Figures 5 and 6). At the pupa stage for 13 °C there were no survivors (Figure 7).

These results for developmental threshold and degree-days were similar to those of Dr. Buckingham's *H. pakistanae* study. His study also showed that the speed of development of *H. pakistanae* increased in direct relation to temperature (Buckingham and Okrah, in preparation).

Number of leaves damaged

Feeding rate was apparently inversely proportional to temperature with a higher feeding rate occurring at lower temperature. For example, at 13 and 15 °C an average of 9 to 10 were leaves damaged, for 33 and 35 °C an average of approximately 6.5 leaves were damaged (Figure 8). In the early stages of the experiment for 13 and 15 °C it was noted, on an average, approximately 52 and 69 leaves were damaged, respectively, as compared to 9 and 10 as the experiment continued to run an average of 102 days for 13 and 15 °C. This could explain why in Figure 8 at 20 °C an average of 11 leaves were damaged.

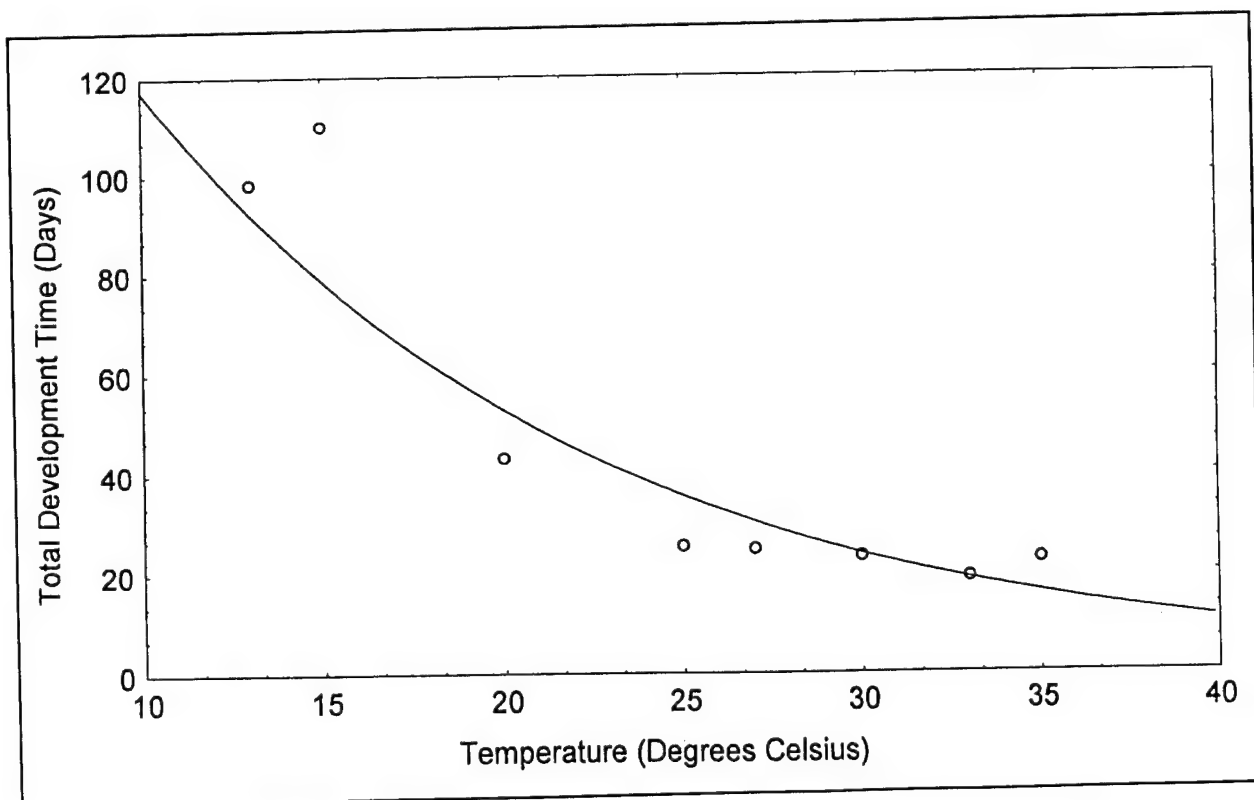


Figure 2. Number of days of overall development from egg to pupa

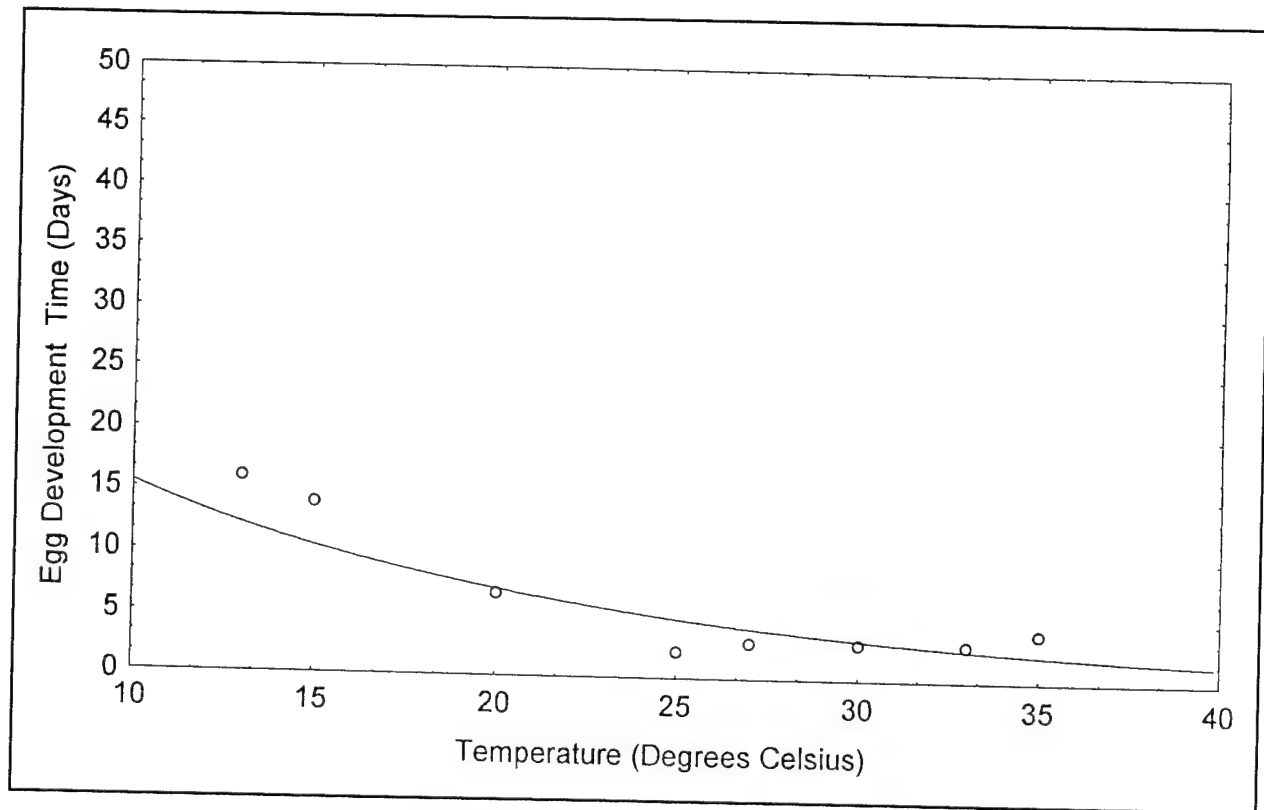


Figure 3. Number of days for egg development at 13, 15, 20, 25, 27, 30, 32.5, and 35 °C

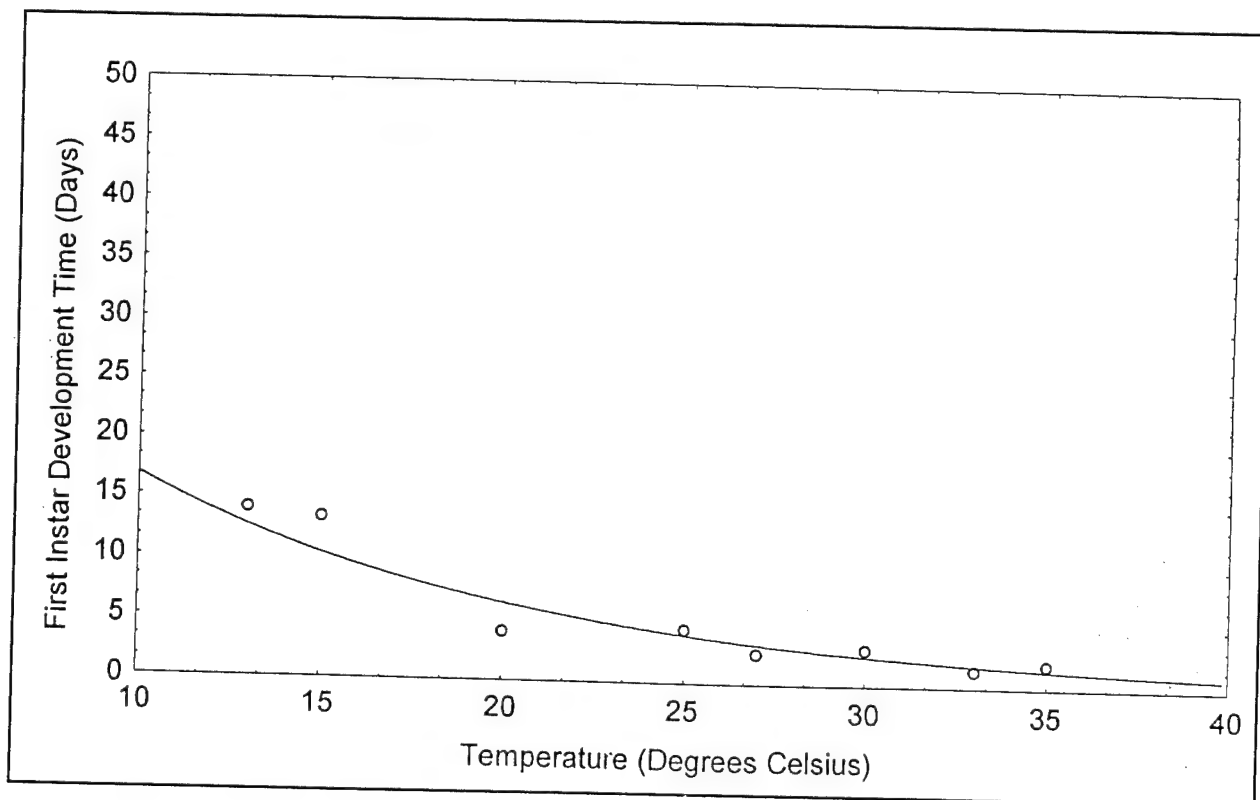


Figure 4. Number of days for first instar development at 13, 15, 20, 25, 27, 30, 32.5, and 35 °C

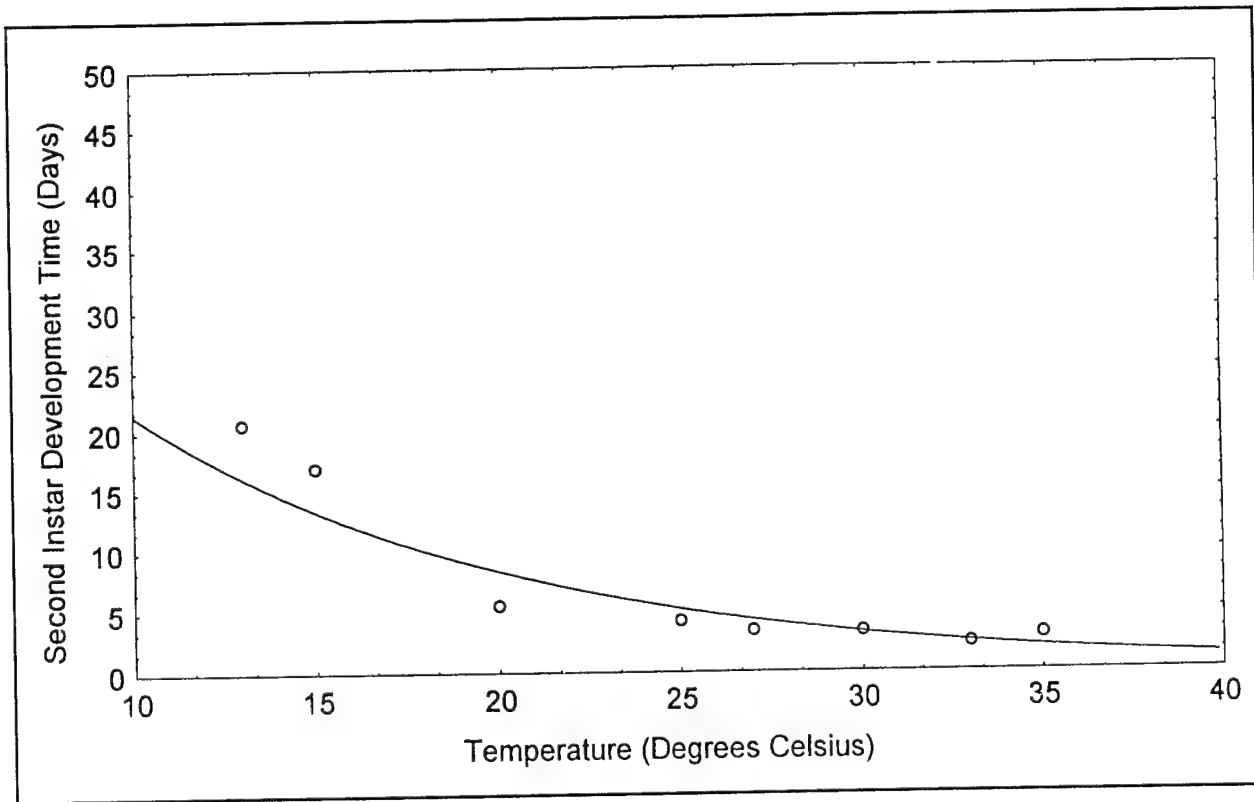


Figure 5. Number of days for second instar development at 13, 15, 20, 25, 27, 30, 32.5, and 35 °C

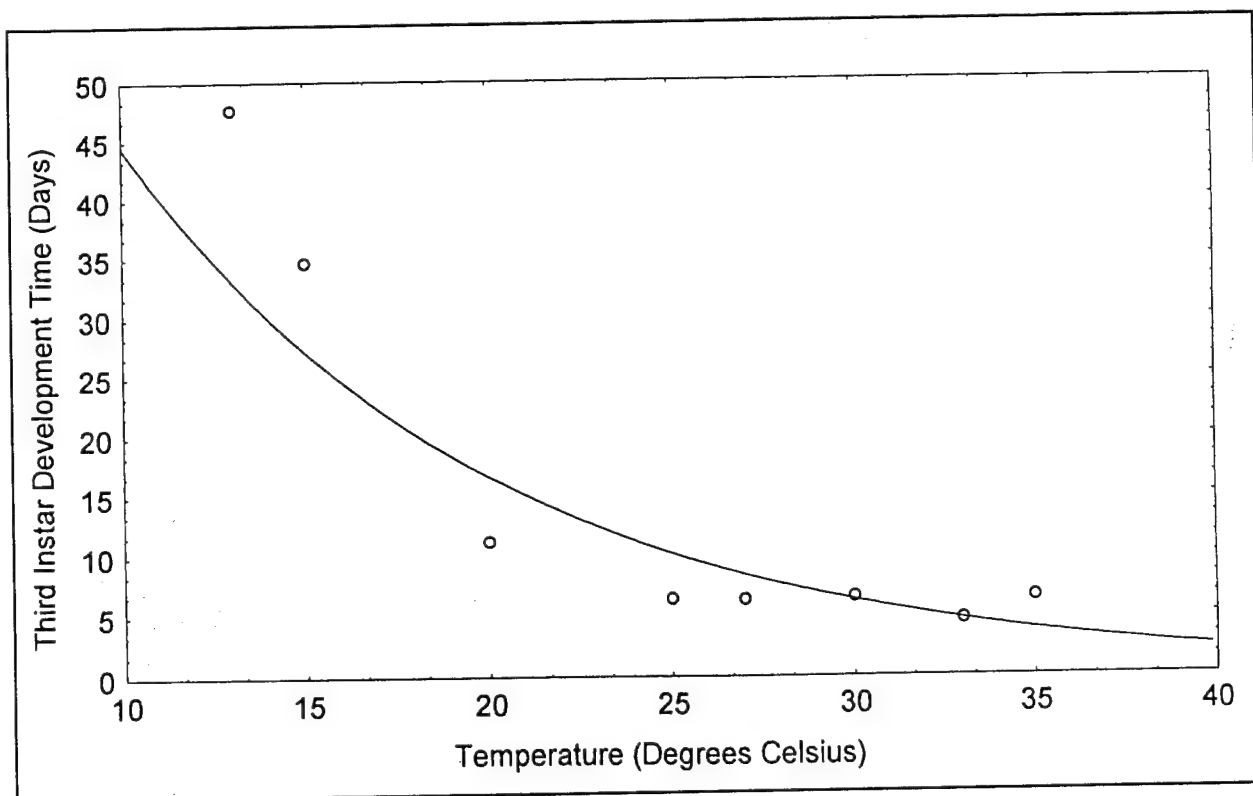


Figure 6. Number of days for third instar development at 13, 15, 20, 25, 27, 30, 32.5, and 35 °C

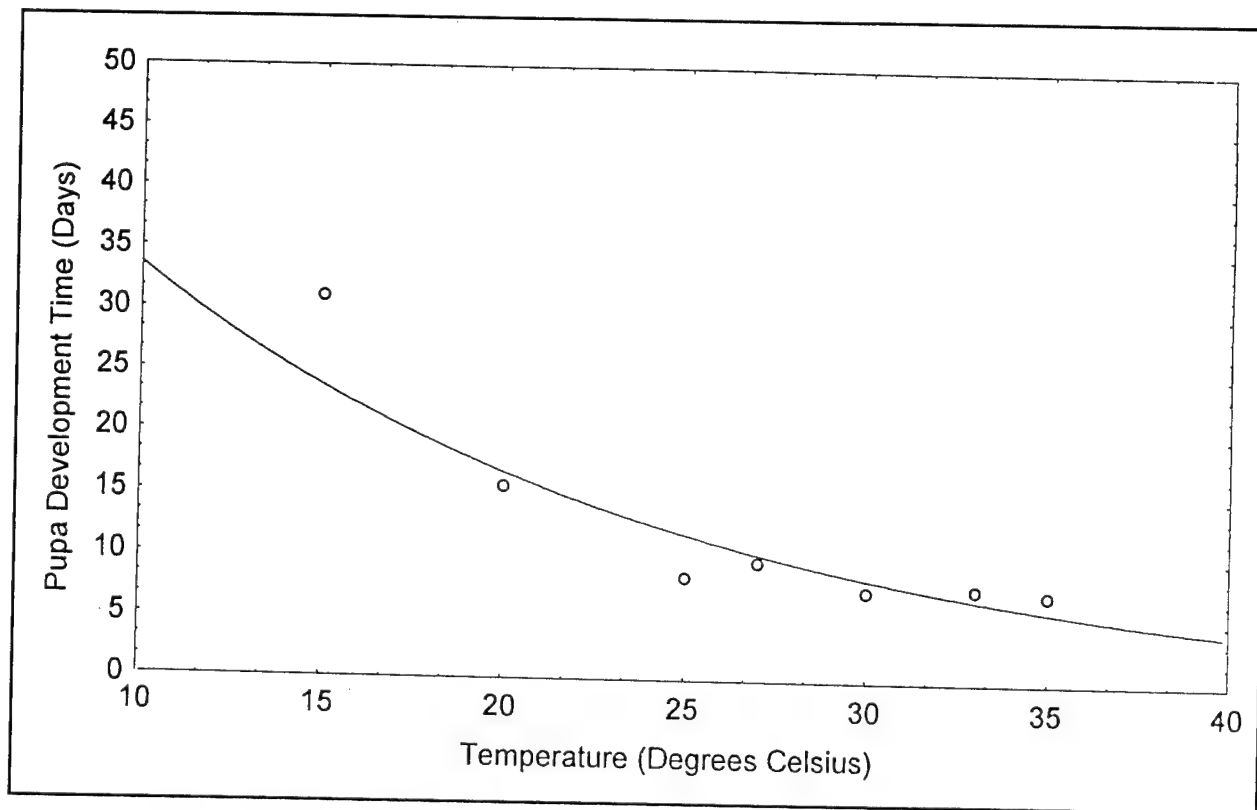


Figure 7. Number of days for pupa development at 13, 15, 20, 25, 27, 30, 32.5, and 35 °C.
(Note that no pupa survived at 13 °C)

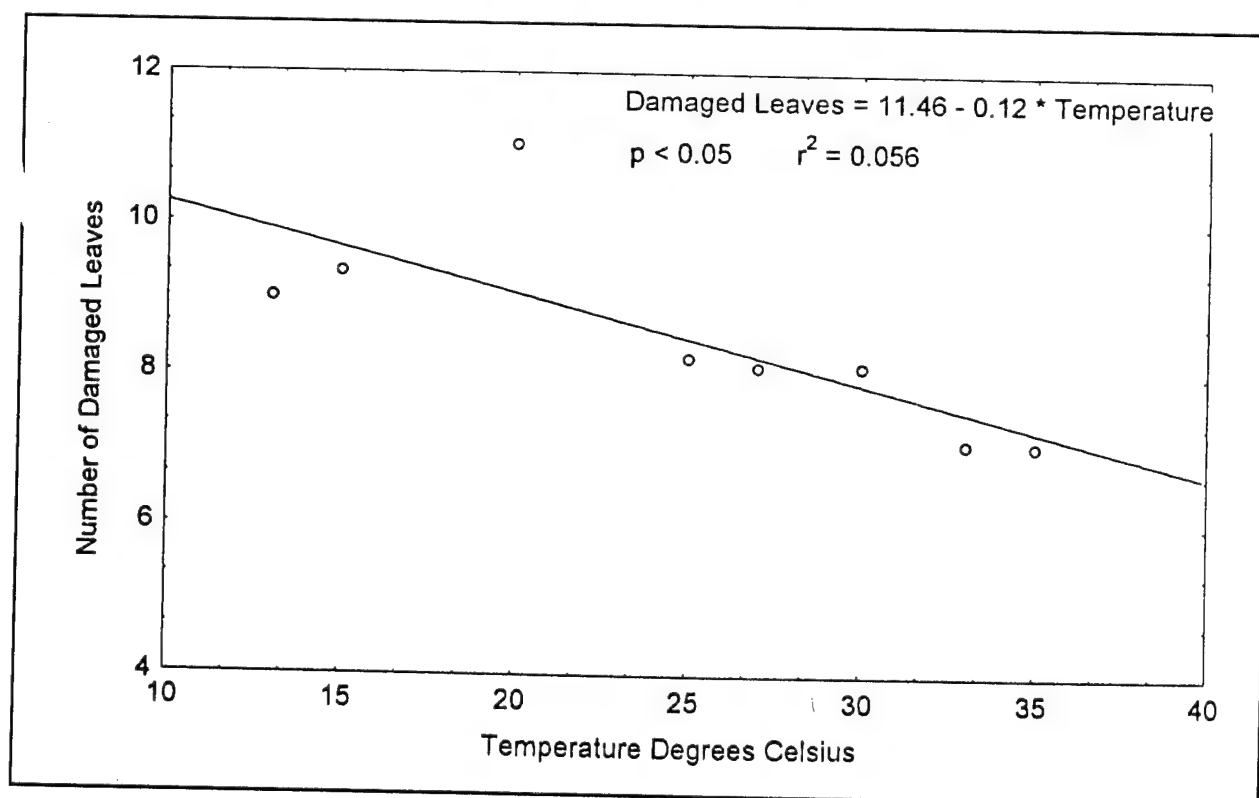


Figure 8. Number of leaves damaged by *H. pakistanae* larval stages

Percent survival

Survival rate was not directly correlated with temperature. We found at 13 and 15 °C, percent survivals were 20 and 10 percent, respectively (Figure 9a). For 20 and 25 °C percent survivals were 56 and 84 percent, (Figure 9b). At 27 and 30 °C, percent survivals were 40 and 48 (Figure 9c), and for 32.5 and 35 °C, percent survivals were 75 and 33 percent (Figure 9d). Greater mortality occurred in first and second instars for all test temperatures.

Cold temperature study

Out of the three different species tested in the cold temperature study *H. balciunasi* had the lowest percentage emergence for all the life stages for the control (0 week), and also for week 1 treatment (1 week in the cold temperature chamber at 6 °C). *H. pakistanae* (India strain) appeared to do better in the control week 0 then the others species tested for all life stages. *H. pakistanae* (China strain) did not do any better then the India strain of *H. pakistanae* in the control week, but in week 1 of exposure time to cold temperature there were only two life stages survivals and they were third instar and pupa. There was approximately 65 percent emergence of pupa for the China strain of *pakistanae*, and approximately 12 percent for the third instar. For exposure time of 2, 4, and 8 weeks there were no emergences for all three species tested (Figure 10).

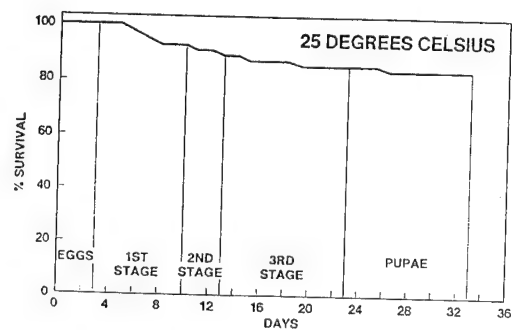
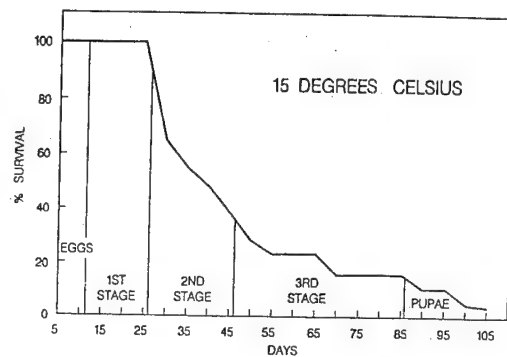
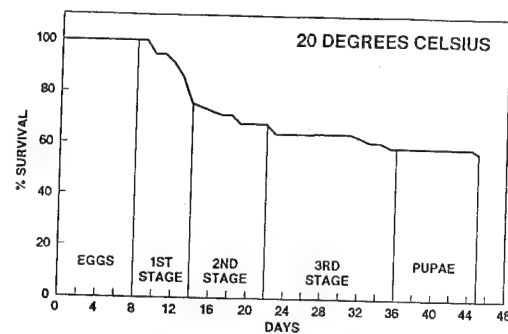
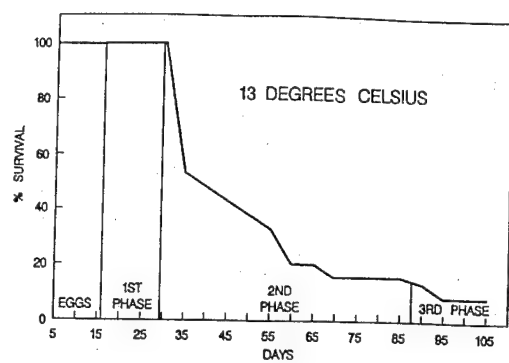
What we found in the lab also is what happened in the field to some extent. As shown in Figure 11, there was some survival at lower temperatures, more so than what we found in the lab.

It is not only important to establish an agent, but also to maintain establishment of biocontrol agents in the field. This is one reason laboratory experiments are important in aiding in that goal. Laboratory experiments such as the ones discussed in this paper will

provide information that could aid in field experiments. For future plans we would like to continue cold temperature studies and examine feeding patterns more closely by looking at the different types of hydrilla the insect feed on.

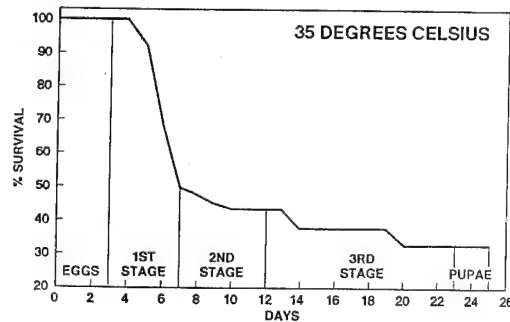
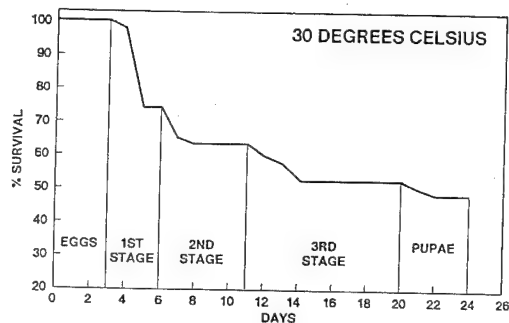
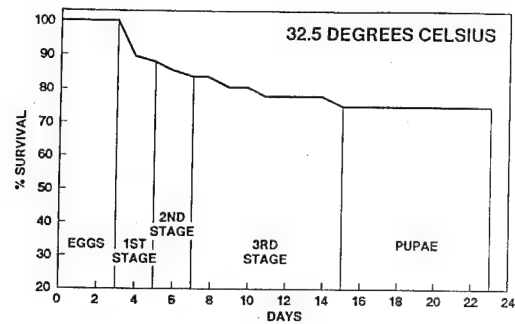
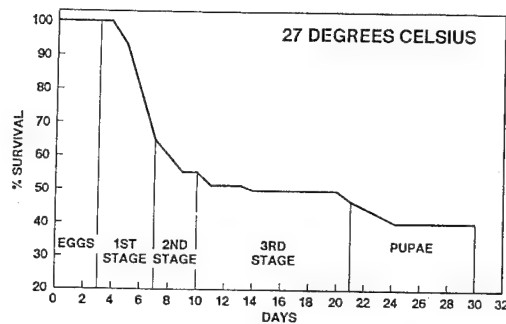
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a.

b.



c.

d.

Figure 9. Percent survival from egg to adult for all temperatures tested

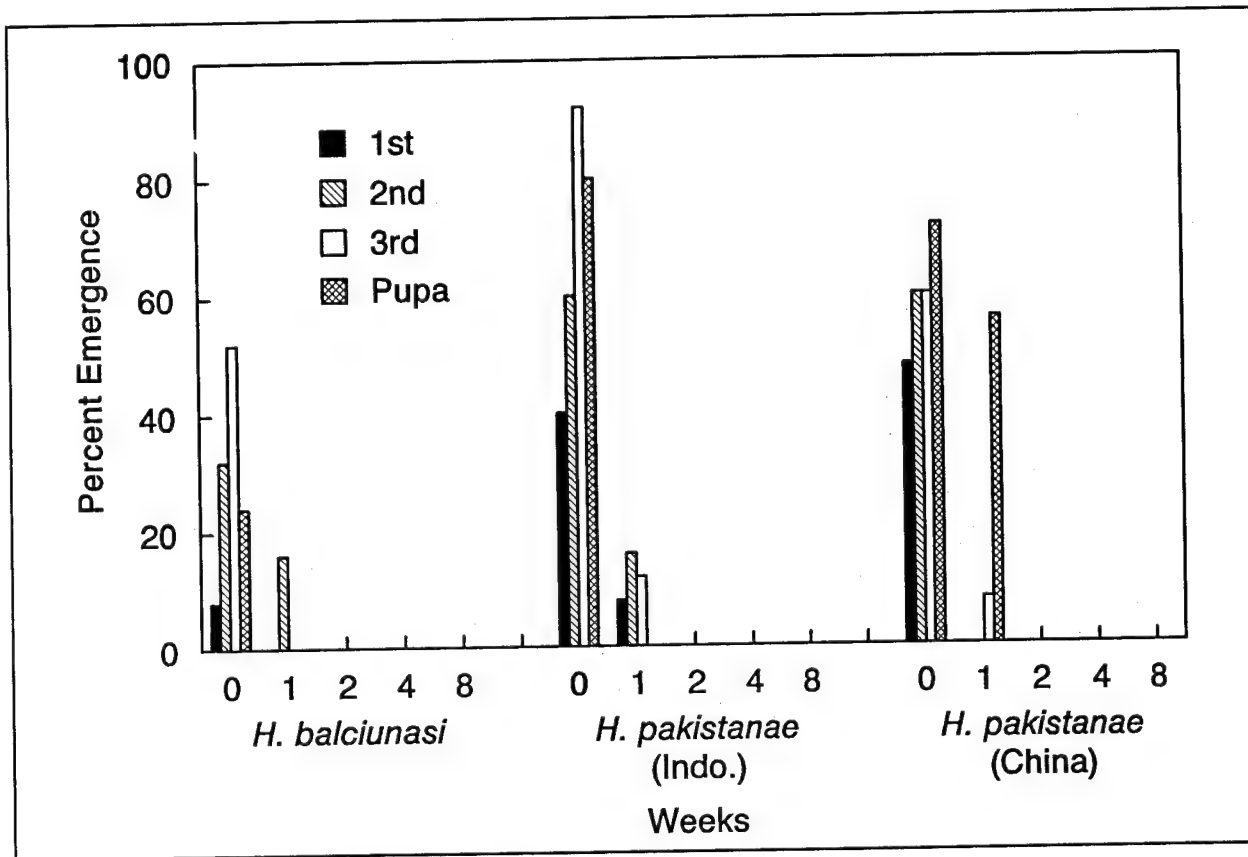


Figure 10. Percent emergence for all three species tested

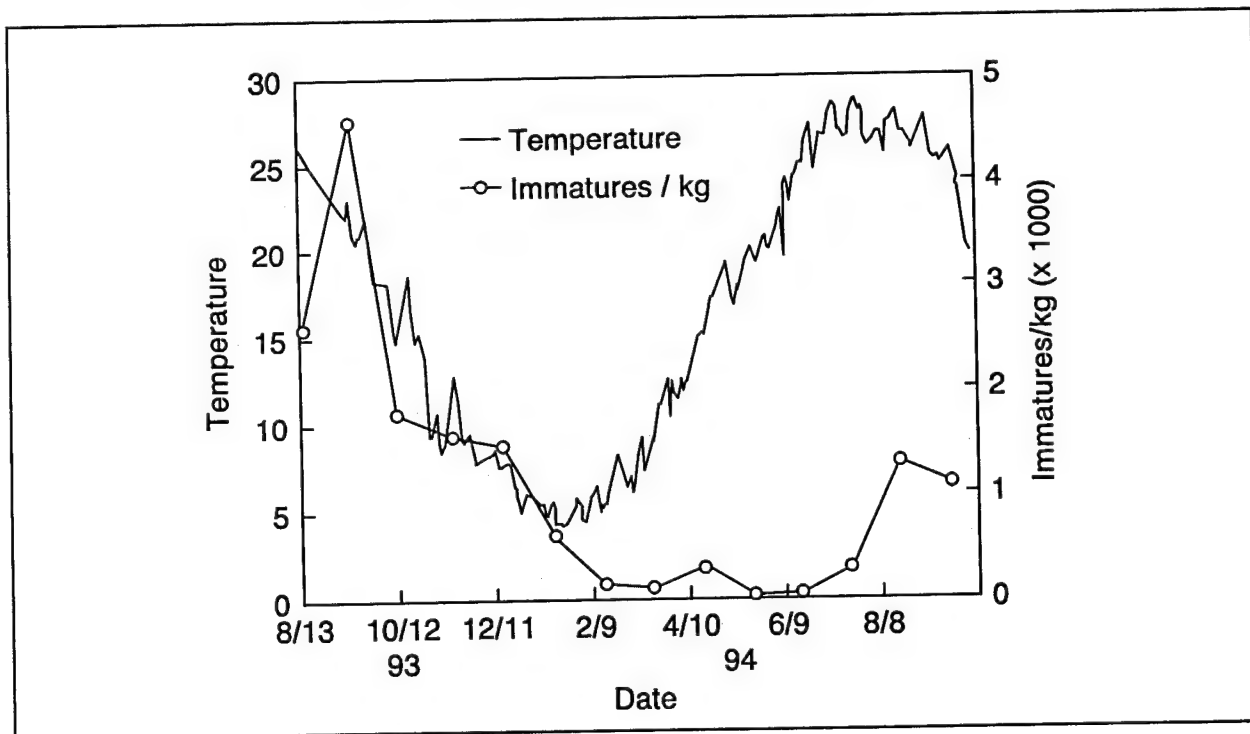


Figure 11. Populations of immature/kg that have been released in the ponds at Muscle Shoals

The Search for Natural Enemies of *Trapa*

by
Robert W. Pemberton¹

This is a status report of the project to discover and define the natural enemies of *Trapa natans* L. and related *Trapa* species. The goal of the project is to locate insects and diseases that have promise to control horned water chestnut or water caltrop (*T. natans*).

The Problem

Horned water chestnut is an aquatic weed that was first observed to be growing "luxuriantly" in Sander's Lake, Schenectady, NY, in 1884 (Wibbe 1886). The plant subsequently spread to many locations in the Northeast where infestations limit navigation, recreation, and the presence of more desirable vegetation (Bickley and Cory 1955; Bogucki, Gruendling, and Madden 1980; Madsen 1993). Biological control is thought to have potential for the management of *T. natans* because this methodology has proved very useful in controlling other water weeds (Center, Cofrancesco, and Balciunas 1989), and also because of the plant's biological characteristics and taxonomic isolation in the United States (Pemberton 1994).

Past Research

Surveys for the natural enemies of *T. natans* and related *Trapa* species were made in Northeast Asia in 1992 and 1993 (Pemberton 1994). Surveys in Japan were made in Hokkaido, the central area of Honshu, and the northern part of Kyushu. In China, surveys were made in Heilongjiang Province, the Beijing area counties, and the lower Yangtze River Delta. In the Russian Far East, the Slavanka and Hinkanski areas of

the Amur River in Khabarovsk Territory were explored. In South Korea, surveys were made in many parts of the country, and in addition, several populations of *T. japonica*, the most common *Trapa* species in the country, were regularly monitored for natural enemy activity and damage.

The most abundant and damaging insect found was the leaf beetle *Galerucella birmanica* Jacoby (= *G. nipponicus* (Walker)). The beetle was abundant in all areas except Russia and Hokkaido. It is a major pest of *Trapa* spp. that are cultivated in the Yangtze River Delta of China for their edible nuts. The beetle also caused the destruction of some of the monitored *T. japonicus* populations in South Korea. Unfortunately, it has host plants in families unrelated to the Trapaceae. This broad host range probably would preclude the beetle's use as a biological control agent of horned water chestnut in the United States.

Pyralid moths with aquatic caterpillars were the next most important herbivores of *Trapa* spp. One, *Nymphula interruptalis* (Pryer), fed upon and destroyed the central buds of many of the monitored plants in South Korea. This moth and the others found on the surveys are reported in the literature to be pests of rice and many other plants.

Two weevils in the genus *Nanophyes* were found attacking *Trapa* spp. Both appeared to be very narrow specialists but did little damage to the plants they attacked. These small weevils feed within the petiole floats of the leaves and no more than one small weevil per leaf was noted. Even when every leaf was attacked, the plants appeared to be normal.

¹ U.S. Department of Agriculture, Agricultural Research Service, Aquatic Plant Management Laboratory, Fort Lauderdale, FL.

An assortment of other generalist insects and diseases was encountered. The surveys in Northeast Asia found some insects that were very damaging but too polyphagous, and others that appeared to be very specific but were not damaging. Even though the plants in many populations were quite injured by insect feeding, no promising biological agents were located.

The surveys found that the insect herbivore fauna was very similar over most of the very large region examined. In addition, no new natural enemy species were located during the second year of surveying, even though the area of search was broadened. This is not to say that all of the herbivores of *Trapa* species occurring in Northeast Asia have been found. It does suggest, however, that the chances of finding different and hopefully more useful agents may increase if new regions are explored.

Future Research

Trapa species are native to both the tropical and warm temperate zones of the Old World (Sculthorpe 1967). The native range of *T. natans* is, in addition to Northeast Asia, Southeast Asia, Central Asia, Africa, and Europe. In Southeast Asia, it is known (as *T. bicornis* Osbeck) from Cambodia, Java, and Thailand (Burkill; 1935, Backer and Bakhuizen Van Den Brink 1963; Cantelo 1965). The plant is reported from India, Iran, and Turkey (Hooker 1879, Cook et al. 1974, Davis 1972). In Africa, it is recorded from the upper Nile and Nigeria (Oliver 1871; Hutchinson and Dalziel 1954). In Europe, the plant ranges from Spain east to Greece in the Mediterranean; northeast through France, Germany to the Baltic area of the former Soviet Union; and east through Central and Eastern Europe to Russia (Tutin et al. 1968).

Where in this vast area should the next surveys be made? The tropical parts of the range are not likely to have enemies adapted to the cold northeastern parts of the United States where *T. natans* is currently a problem. The occurrence of *T. natans* and other *Trapa* species in warm areas suggests they could do well in the warmer parts of the United States.

If *T. natans* spreads or is introduced to the southeastern United States or to Florida and becomes a problem, the warmer parts of its native range could become important sources of natural enemies. The fruits of *Trapa bicornis* are imported and sold for food in Chinese markets in the United States, and *Trapa* species have been marketed in the United States as ornamentals (Porterfield 1938).

In South Central Asia, the northern cooler parts of India and Pakistan are of interest. Weevils, including *Bagous* spp., have been used as biological control agents of water weeds (Room et al. 1981; Center, Cofrancesco, and Balciunas 1989) and a *Bagous* weevil (*B. trapae* Parshad) has been recorded from *Trapa bispinosa* in India (Parshad 1960). The occurrence of *T. natans* in this area is apparently limited, and Kashmir, the area with the best known populations, is presently unsafe to visit. *Galerucella birmanica* is known to be a pest of cultivated *Trapa* in India, like it is in China, which suggests at least some faunal similarity between the regions.

Trapa natans appears to be more common in Europe, occurring at least to some degree in all of the countries of the central and southern areas, although few exact localities are known. There is also a *Bagous* weevil reported from *T. natans* in northern Italy. Since Europe represents the western extreme of *T. natans*' native temperate range, one could expect greater differences in the natural enemies of the plant in Europe than would occur in regions closer to Northeast Asia, such as Central Asia. Europe, therefore, seems like the best region to search for new natural enemies of *T. natans*.

In 1995, the work plan is to combine a museum study and a field survey in Europe. The British Museum, Paris Museum, and St. Petersburg Zoological Institute will be visited to examine herbarium material of *Trapa* spp. in order to obtain locality records for Europe and Central Asia, including India. Insect systematists and plant pathologists will be consulted to learn of insects and diseases affecting *Trapa* spp. In a related effort, I will try to develop a cooperative USDA-ARS—Russian Zoological

Institute project to examine the Russian literature and museum material to learn of Russian insects affecting aquatic weeds that are problems in the United States. Based on the *Trapa* locality data obtained, a field survey will be designed and conducted. Northern Italy, the area from which the *Bagous* was reported, and southern France, where USDA-ARS has its European Biological Control Laboratory (which can provide survey support) will probably be included. Depending on the locality records obtained for India, as well as the political situation next year, a survey may be made there in 1996.

Acknowledgments

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Technology Transfer Activities for Biological Control

by
Michael J. Grodowitz¹

Introduction

The use of biological control is a viable, alternative strategy for the management of several species of problem aquatic plants. The biocontrol program began in 1959 with an informal cooperative agreement between the USDA and the USACE. Since that time, the biocontrol program has grown tremendously with a total of 12 insect agents released for the management of alligatorweed, waterhyacinth, waterlettuce, and hydrilla. The majority of these agents have become established in as many as 11 different states. More significantly, this technology has been exported to least 20, if not closer to 30, other countries, worldwide. Effective biocontrol requires close cooperative ties with many different agencies and institutions. Toward this goal, the aquatic plant biocontrol program has had close cooperative ties with at least three major Federal agencies, five state institutions, numerous overseas laboratories, and many local and academic establishments. In addition, the need for this technology is still expanding. Additional problem plants of both aquatic and wetland habitats have been recognized as possible biocontrol targets. These include Eurasian watermilfoil, water chestnut, purple loosestrife, Brazilian pepper, Chinese tallow, among others. Biocontrol represents an environmentally compatible, long-term management strategy and, when used correctly, can be a productive and vital part of most existing aquatic plant management programs.

As with any management technique biocontrol requires an extensive base of knowledge and hands-on experience to be used correctly and efficiently. Unfortunately, most operational personnel know little concerning

the correct and active use of biocontrol technology. A typical perception of biocontrol is that it is a rather passive technology where agents are released at field sites, mainly by researchers, and allowed to grow and feed with little managing input from man. Such a perception of biocontrol has flourished because of a real and perceived lack of information concerning the correct and active use of biocontrol technology summary information that is specifically geared to personnel at the operational level.

Since the beginning, biocontrol researchers have recognized the need for appropriate technology transfer mechanisms. Many traditional technology transfer tools have been used including technical reports and publication in scientific journals, the development of video presentations, and publication of articles in the popular literature. Over the last several years, however, several new approaches to technology transfer in the biocontrol field have been developed. These include a variety of old and new procedures to transfer information to field personnel and include: (1) the development of a short course that details general and specific concepts on the use of biocontrol, (2) formal publication of a USDA - ARS and Florida Department of Natural Resources manual to serve as a primer on the use of biocontrol, (3) formation of cooperative extension-like activities for personnel at District and state agencies, and (4) development of a computer-based information system that details the history and use of biocontrol for aquatic plant management. The following report is a short review and current status of these new and upcoming technology transfer approaches.

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Short Course

Since 1992, biocontrol researchers have been developing a short course that deals with general and specific issues on the use of biocontrol technology. The course was developed with funds provided by the APOSC, Jacksonville District, and the APCRP. Authors of the short course include Dr. M. J. Grodowitz and Dr. A. F. Cofrancesco from the WES, Dr. G. Buckingham and Dr. T. Center from the ARS, and Dr. W. Johnson from Nicholls State University.

The course covers a wide range of topics. These include a brief background on ecological theory and population biology, historical aspects on the use of biocontrol, how biocontrol is achieved, advantages and disadvantages of using biocontrol, the active use of biocontrol in existing aquatic plant management programs, as well as detailed information on all of the introduced agents and commonly encountered native herbivores.

Students who participate in the course receive a complete set of the course notes. This allows students to listen to the presentation without the need to record large quantities of information. The course is very graphical in nature containing over 300 photographs, graphs, and illustrations of various aspects of biocontrol.

The first class is to be presented by Jacksonville District personnel during March 1995. The number of classes given will depend on interest generated by field personnel. The first year will serve as a field review of the course allowing class participants the chance to provide suggestions on how the course can be improved in later years. If you are interested in participating in this course please feel free to contact Dr. William Zattau (904/232-2215).

Insect Manual

Another technology transfer tool that is nearing completion is the development of a manual to serve as a primer on the use of biocontrol. The manual's introductory sec-

tion is a relatively detailed introduction or primer on the use of biocontrol that contains much information on the history, ecological background, and use of biocontrol. This introductory section is a summary of information that is presented in the short course mentioned previously. The main portion of the manual will contain full color plates and detailed textual information on all of the introduced insect agents as well as information on commonly encountered native herbivores associated with at least 17 different aquatic and wetland plant species. The manual will serve as an introductory text and field guide for operational personnel on the use of biocontrol for aquatic plant management. This manual is based largely on a field guide jointly developed by USDA - ARS and Florida Department of Natural Resources (FDNR) researchers. However, the USDA, ARS, and FDNR field guide was produced in only limited quantities for a fairly restricted distribution thus the need for a formally published manual.

Cooperative Extension Activities

The use of cooperative extension-like activities for state and District personnel is a growing part of our technology transfer approach. Cooperative extension activities are varied but mainly involve close communication and cooperation between researchers and operational personnel on specific aquatic plant biocontrol uses and problems. Cooperative extension activities usually involve working closely with operational personnel that are in the process or want to apply biocontrol technology to site-specific problems. While such activities have been ongoing since the inception of the biocontrol program, these activities have recently taken on a more organized approach. Over the last several years, with funding provided mainly by the Galveston District and the APCRP, biocontrol researchers from WES have worked closely and cooperatively with personnel from the Texas Parks and Wildlife Department (TPWD) on a variety of aquatic plant biocontrol topics. These have included hydrilla, waterhyacinth, and water-lettuce infestations involving the release,

monitoring, and establishment of four different insect agents. Cooperative extension activities are an important part of our technology transfer.

Information System

A new technology transfer tool that has recently been developed involves the use of computer-based information systems. These systems allow for rapid and efficient access to a wide variety of information on the use of biocontrol. Two inter-connected information systems have been developed and are closely tied to the short course discussed previously. These systems allow for rapid and relatively easy identification of the introduced and native herbivores as well as their associated damage. These systems include information on the herbivores that feed on the four primary problem plants; alligatorweed, waterhyacinth, hydrilla, and waterlettuce. These systems use a highly interactive graphical interface that closely simulates the interaction between nontechnical personnel and experts in the biocontrol field. Information that can be accessed includes identification strategies, summary information from that which is presented in the short course, introduction to insect and invertebrate morphology used for identification, as well as information on the active use of biocontrol in existing aquatic plant programs. This information is presented using large quantities of textual information as well as over 250 different photographic quality images and illustrations on the agents and their associated damage. The systems are highly portable, able to be run on any PC-based machine running Windows 3.1 in 256 color video mode and 20 megabytes of free hard disk space.

These systems are in the process of being reviewed during FY95 together with the scheduled review of the short course. Participants in the course will receive a complete disk copy of the information systems as well as detailed instruction on their use. Computer-based technology transfer allows for the transfer of large quantities of information rapidly with lower costs for future updates and revisions. Compilation of information in a com-

puter format is substantially less expensive than more traditional means, such as books and technical reports, especially considering the costs associated with publishing full color photographs.

The system on the herbivores of aquatic plants is also being expanded to include all of the species included in the soon to be published manual on the use of biocontrol. The new system will not only contain the commonly encountered herbivores associated with alligatorweed, waterhyacinth, hydrilla, and waterlettuce but also 14 additional aquatic and wetland plant species such as duckweed, smartweed, Eurasian watermilfoil, cattail, etc. Due the large amounts of information contained in this system these may be distributed on CD-ROM instead of the more traditional floppy disks.

The future of computer-based technology transfer is great. Other aspects of aquatic plant management can easily be incorporated into these systems. For example, a recently developed prototype information system developed for the Department of Defense's Strategic Environmental Research Development Program (SERDP) incorporates information access not only for biocontrol technology but also for plant identification and chemical application as well. A similar system is also being developed for information access on zebra mussel basic biology, sampling protocols, risk assessment for various facilities, and associated control strategies. Future add-ons for aquatic plant control can include a wide variety of information including aquatic plant databases, computer simulations, short video applications, etc. The only limitation is funding and your imagination.

Summary

Technology transfer activities are an important and integral component of any research program. Standard technology transfer tools such as technical reports and various other bound publications are but one important approach. Other technology transfer areas can include the development of short courses,

field guides and manuals, as well as cooperative extension activities. Another growing field is the application of computer technology

in the development of highly interactive information systems which allow for rapid and efficient information access.

Ecology of Aquatic Plant Technology

Effects of Submersed Macrophytes on Water Quality: A Synopsis

by

John W. Barko¹ and William F. James¹

During the 1970's, accelerated eutrophication of freshwater systems due to excessive phosphorus loadings in both North America and Europe lead to interest in the role of rooted submersed macrophytes in the phosphorus economy of the aquatic environment. At that time it was unclear whether submersed macrophytes functioned as sources or sinks for phosphorus. Given potential access by these plants to nutrients in both the water column (foliar uptake) and sediments (root uptake), it was necessary to evaluate quantitatively nutrient source-sink relationships. Using a mass balance approach in controlled environmental settings at WES, we evaluated the nutritional ecology of several species of submersed macrophytes. During the late 1970's and early 1980's these investigations, in combination with related efforts by others, demonstrated the significance of sediment as the primary source of major nutrients for uptake by rooted submersed macrophytes. As importantly, these investigations indicated also that submersed macrophytes operated as potential sources of nutrients during periods of senescence. These investigations constituted the first significant efforts directed toward better understanding the effects of submersed macrophytes on eutrophication. Subsequent studies conducted in the APCRP during the late 1980's and early 1990's have further elucidated the effects of submersed aquatic macrophytes on water quality. The purpose of this article is to provide a synopsis of major findings of these subsequent studies.

Macrophytes on Sediment Phosphorus Release

The process of photosynthesis in submersed macrophytes results in oftentimes dramatic increases in the pH of the surrounding water column. For example, studies conducted with the U.S. Geological Survey in *Hydrilla* beds in the Potomac River revealed diel changes in pH between values of about 7.0 and 10.0. Since pH values are logarithmic, these changes indicate large (nearly one thousand-fold) variations daily in hydroxyl and hydrogen ion concentrations. From concurrent studies conducted at Eau Galle reservoir in Wisconsin, it became apparent that elevated pH beyond about 9.0 can result in significant increases in rates of phosphorus release from surficial sediments. In simple terms, the mechanism for release appears to involve chemical exchange of hydroxyl ions for phosphate ions in sediment.

Phosphorus released from sediments under conditions of elevated pH can account for a significant portion of the total mass of this element loaded internally in aquatic systems. For example, we demonstrated that about 20 percent of the total seasonal internal phosphorus load into Eau Galle reservoir derived from littoral sediments. Phosphorus released from littoral sediments (i.e., within macrophyte beds) tends to be transported directly into the upper mixed layer of lakes and reservoirs (see below). Thus, contributions to the phosphorus economy of algal communities in surface

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waters can be significant. The seasonal periodicity of internal phosphorus loadings into the upper mixed layer of Eau Galle reservoir appears to influence not only the nutrient budget, but phytoplankton productivity and the vertical migratory behavior of phytoplankton populations as well.

Macrophytes on Hydraulic Circulation

On a daily basis, shallow near-shore regions of aquatic systems typically heat and cool more rapidly than deep open-water regions, due primarily to differences in mixed volume (see article by Smith and James, this proceedings). The presence of submersed macrophytes in shallow regions contributes to the development of thermal gradients in both the vertical and lateral planes, as foliage near the water surface converts solar irradiance to heat. Thermal gradients give rise to density gradients that promote hydraulic circulation. Implications of hydraulic circulation driven by convection are potentially far-reaching, since dissolved constituents (e.g., phosphorus released from sediments, see above) can be moved with water. Dissolved constituents may include contaminants or herbicides in addition to nutrients. As emphasized above, hydraulic transport from the littoral zone can contribute significantly to pelagic nutrient budgets. With herbicides, information on the periodicity of hydraulic transport can be of enormous value in maximizing both the efficiency and effectiveness of treatment applications (see articles in Chemical Control Technology section, this proceedings).

In Eau Galle Reservoir in Wisconsin and Guntersville Reservoir in Alabama, dye studies (see article by Smith and James, this proceedings) have been conducted for several years in combination with close-interval thermal monitoring in an attempt to evaluate the seasonal dynamics of convective circulation. Owing to the eutrophic nature of these impoundments, studies have focused primarily on phosphorus transport. However, the results obtained from these reservoir studies

apply to many dissolved constituents. These results indicate the potential significance of shallow water macrophyte beds in affecting chemical budgets in aquatic systems.

Macrophytes on Sediment Resuspension

In addition to effects on soluble constituents of the water column in aquatic systems, submersed macrophytes also play an important role in mediating the resuspension and transport of sediment and associated particulate constituents. Sediment resuspension and discharge of sediment downstream in Marsh Lake (Minnesota) were examined during 1991 and 1992 under a variety of wind conditions (see James and Barko, this proceedings). Based on a theoretical wave model, nearly the entire sediment surface area of this reservoir (81 to 100 percent) can be disturbed by wave activity at wind velocities as low as 15 km/hr blowing from any direction. However, as an apparent result of dense submersed macrophyte beds that in 1991 covered nearly the entire lake, measured sediment resuspension was much less frequent than expected from wave theory.

Critical thresholds of wind velocity required to resuspend sediment in Marsh Lake were much higher in 1991 than in 1992 when plants were essentially absent. The presence of dense submersed macrophyte populations in 1991 resulted in a much lower frequency of resuspension events than in 1992. In addition, discharge of resuspended sediment downstream was much less in 1991 when submersed macrophytes were abundant than in 1992 when macrophytes were absent.

In Marsh Lake submersed macrophyte populations appear to significantly influence water quality conditions (e.g., chlorophyll concentrations and turbidity) by mediating sediment resuspension. Reduced discharges of sediment from this system appear to result in water quality improvements downstream as well.

Conclusions and Recommendations

From the foregoing it is apparent that submersed aquatic macrophytes, through a variety of mechanisms, can have important influences on water quality conditions in lakes and reservoirs. The significance of macrophyte influences on water quality can be expected to vary with climate, basin geomorphology, macrophyte density, and species composition. In systems where water quality is of significant concern, aquatic plant management practices should be devised and implemented with consideration for water quality effects.

Aquatic plant management needs to be viewed holistically, since the goal of aquatic plant management in many cases may be an increase, rather than a decrease, in the distribution of submersed aquatic macrophytes. Results of studies conducted in Marsh Lake (see above) suggest that the development and maintenance of stands of submersed aquatic macrophytes may be an effective management tool for limiting wind-driven sediment resuspension and sediment discharge in shallow impoundments and lakes. Thus, macrophyte growth in Marsh Lake should be encouraged, perhaps by engineering means (e.g., construc-

tion of artificial islands, provision of sediment retention basins, etc.), rather than discouraged.

The CE is presently engaged in major efforts directed toward habitat improvement through projects designed to encourage expansion of macrophyte communities in backwater areas of the Upper Mississippi River. Unfortunately, habitat improvement projects are at present based on numerous untested assumptions regarding linkages among aquatic macrophytes, water quality, and habitat value. These assumptions need to be evaluated in future ecological study efforts. As demonstrated in the APCRP-sponsored studies summarized above, direct linkages exist between macrophytes and various facets of water quality. However, relationships between water quality conditions affected by macrophytes and the use of macrophyte beds by the biota of lakes and reservoirs have not yet been clearly defined.

Acknowledgments

The information summarized herein derives from the efforts of a large number of investigators who have over the years contributed substantially to progress in the Ecological Technology Area of the APCRP.

Effects of Submersed Macrophytes on Sediment Resuspension in Marsh Lake, Minnesota

by
William F. James¹ and John W. Barko¹

Introduction

Shallow lakes and reservoirs are often dominated by wind-induced sediment resuspension leading to sediment-related water quality problems, such as enhanced nutrient recycling and reduced water clarity (Dillon, Evans, and Molot 1990; Maceina and Soballe 1990; Hellström 1991; Søndergaard, Kristensen, and Jeppesen 1992). By reducing sediment resuspension, wetland and submersed aquatic vegetation can have beneficial effects on the water quality of shallow impoundments and lakes. For example, Dieter (1990) showed that sediment resuspension was reduced in regions colonized by emergent vegetation in Sand Lake, SD. Similarly, James and Barko (1990) demonstrated that submersed vegetation reduced sediment erosion in the littoral zone of Eau Galle Reservoir, WI. Thus, aquatic vegetation may play an important role in mitigating water quality problems in shallow lakes subjected to frequent winds.

In this study, we examined sediment resuspension in Marsh Lake, MN, and the discharge of sediment from the system to downstream Lac Qui Parle Reservoir, MN. Sediment resuspension was examined under a variety of wind conditions in Marsh Lake to determine critical thresholds of wind velocity that resulted in high concentrations of suspended sediment (i.e., seston) in the water column. Sediment resuspension was examined during a year when submersed aquatic vegetation in Marsh Lake was densely established (1991) and during a year when it was almost completely absent (1992), thus providing information on the influence of aquatic vegetation on sediment dynamics in a shallow reservoir.

Methods

Marsh Lake, a flood-control impoundment located near the headwaters of the Minnesota River in western Minnesota, is large (surface area = 1,862 ha) and very shallow (mean depth = 0.9 m and maximum depth = 1.5 m; Figure 1). Tributaries to Marsh Lake include the Pomme De Terre River, which enters the lake near the dam and outlet structure, and the Minnesota River (Figure 1). The tributaries drain a mostly agricultural watershed. Discharges from Marsh Lake occur over an uncontrolled fixed-crest overflow structure located at an elevation of 285.78 m NGVD. Discharge also occurs through a sluice gate located at 284.26 NGVD. Discharges from Marsh Lake travel approximately 20 km downstream before entering the headwaters of Lac Qui Parle Reservoir.

Sediment loading and resuspension were examined in Marsh Lake during June through September 1991, and during May through September 1992. During the summer and autumn of 1991, the entire reservoir was densely populated by *Potamogeton* sp., with surface coverages >90 percent.² Submersed macrophyte densities diminished greatly in 1992, to the point where plants could not be observed visually in the water column throughout most of the reservoir. Differences in macrophyte coverage were not quantified, since we did not anticipate such a marked contrast in abundance between the two study years. The cause of this macrophyte decline has not been determined.

In 1991, two midchannel stations (0.7 to 0.8 m deep) were established in the lake, with

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² Personal observation, D. Hatfield and K. Bonema.

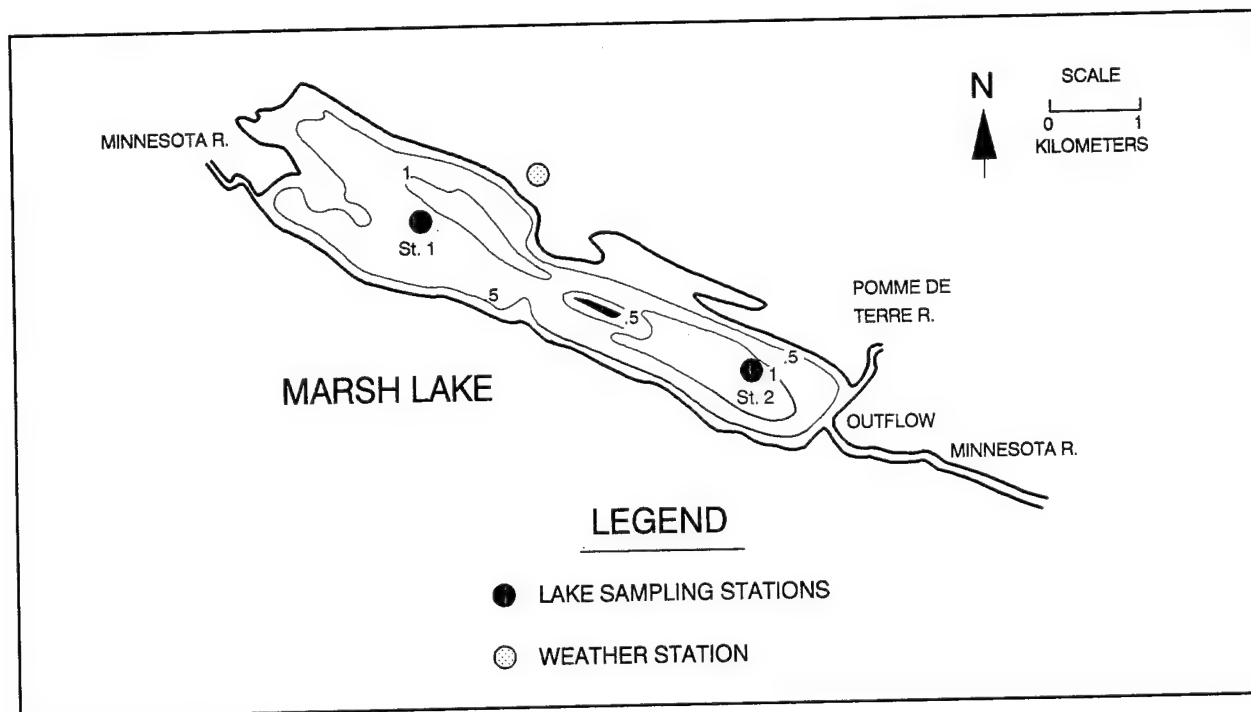


Figure 1. Morphometric map and station locations in Marsh Lake. Depth contours are in meters

water sampling (integrated water column) attempted by boat every Monday, Wednesday, and Friday. However, strong winds and high waves prevented manual collection of water samples on many dates. In 1992, automated water samplers (ISCO) were placed on permanently anchored platforms at these same lake stations (Figure 1). The samplers were programmed to composite three water samples, collected at 8-hr intervals at a depth of 0.3 m below the lake surface, on a daily basis. Thus, water samples could be collected even under severe wind conditions.

River water samples were collected daily (automated water samplers) or weekly (grab samples) at fixed stations located on the Minnesota River upstream of Marsh Lake, on the Pomme De Terre River, and at the outflow of Marsh Lake during both years. Flows were gauged continuously using continuous recording gauges or stage height recorders. The water level of Marsh Lake was measured daily with a recording gauge.

All lake and river samples were analyzed for seston (i.e., total suspended solids), particulate organic matter (POM), and total phosphorus (total P) concentrations. Samples for

seston and POM were filtered onto pre-combusted glass fiber filters. The filters were dried at 105 °C for seston analysis and then combusted at 550 °C for 2 hr for determination of POM (i.e., loss on ignition; APHA 1989). Total P of unfiltered samples was determined colorimetrically on a Technicon Autoanalyzer II following persulfate oxidation (APHA 1989).

Daily external loadings and discharges of seston, POM, and total P in Marsh Lake were calculated by multiplying the daily volumetric flow rate by the measured concentration. Linear interpolation was used to estimate concentrations on dates when water samples were not collected. Retention (kg/d) of these constituents in Marsh Lake was estimated according to the following equation: Retention = External Load - Outflow. The external load represented the sum of daily loading estimates from the Minnesota and Pomme De Terre Rivers while the outflow represented the daily discharge load from Marsh Lake.

A weather station was established on the northern shoreline of Marsh Lake for wind monitoring (Figure 1). The average of data collected at 5-min intervals was recorded

every 15 min to one half hour for wind velocity and direction using an automated data logger (OmniData International). Mean daily wind velocities were computed from these data for comparison with trends in water quality variables, because mean daily velocity reflects sustained winds. Daily wind vectors were calculated as the wind direction weighted with respect to wind velocity and averaged over a 24-hr period. Statistical analyses of the wind data were performed using SAS (1988).

The ability of waves, generated by wind activity, to disturb and resuspend the sediment surface in Marsh Lake was estimated for a variety of wind directions using the wave model developed by Carper and Bachmann (1984). In general, the model calculates wavelengths (L) of surface waves using mathematical wave theory as applied by the U.S. Army Coastal Engineering Research Center (1977), and compares $1/2L$ to the water column depth. If, $1/2L$ is greater than the water column depth, the wave is assumed to touch the lake bottom and resuspend sediment. The wave model assumes that there are no obstructions in the water column such as submersed or emergent aquatic macrophytes that might create drag or redirect currents and wave activity.

Results

During both years, winds blew most frequently out of the southeast (~40 percent) and southwest (~39 percent), followed by winds blowing out of the northwest (18 percent). Relatively low mean daily wind velocities of 5-10 km h^{-1} were most frequent (47 percent)

during both years, followed by greater wind velocities of 10-15 km h^{-1} and 15-20 km h^{-1} , occurring 28 and 11 percent of the time, respectively. Mean daily wind velocities of $<5 \text{ km h}^{-1}$ occurred on only 9 percent of the days during both years. In general, mean daily wind velocities for all directions were $\geq 10 \text{ km h}^{-1}$ on 44 percent of the days and $\geq 20 \text{ km h}^{-1}$ on 5 percent of the days during both years.

Based on wave theory, only 17 to 22 percent of the lake bed was potentially affected by waves at low wind velocities (i.e., $\leq 5 \text{ km/h}$; Table 1). However, at wind velocities of 10 km h^{-1} the percentage of the lake bed affected by waves increased markedly (Table 1), particularly for winds blowing in the direction of maximum effective fetch (i.e., SE and NW winds). Virtually 100 percent of the lake bed was potentially affected by waves at wind velocities of 15 km h^{-1} blowing from any direction. Water sampling stations in Marsh Lake were affected by wave disturbances at critical (theoretical) wind velocities ranging between 10.5 and 15 km h^{-1} , depending on wind direction (Table 2).

In 1991, mean daily wind velocities fluctuated between about 5 and 19 km h^{-1} from June through mid-September (Figure 2a). Peaks in mean daily wind velocity of 19 km h^{-1} occurred in late September (Figure 2a). Wind direction during these periods of high wind velocity was predominantly from the northwest. Seston concentrations were relatively low (i.e., 2 mg/L) in Marsh Lake from June through mid-September 1991 (Figure 2b); however, concentrations of seston increased

Table 1
Percent of the Lake Bed Disturbed by Waves at Various Wind Speeds and Directions

Wind Speed, km/h	Winds Blowing from the			
	NE	SE	SW	NW
5	22	22	17	17
10	49	67	37	75
15	86	95	81	100
20	100	100	100	100

Table 2
Comparison of Effective Fetches and Critical Wind Velocities Required to Resuspend Sediment at Sta 1 and 2 for Various Wind Directions at Nominal Water Column Depths (0.8 m)

Wind Direction	Sta 1		Sta 2	
	Fetch, m	Critical Velocity, km/h	Fetch, m	Critical Velocity, km/h
NE	1,100	15.0	1,150	15.5
SE	1,200	14.5	2,500	11.0
SW	1,100	15.0	1,200	14.5
NW	2,900	10.5	1,900	12.0

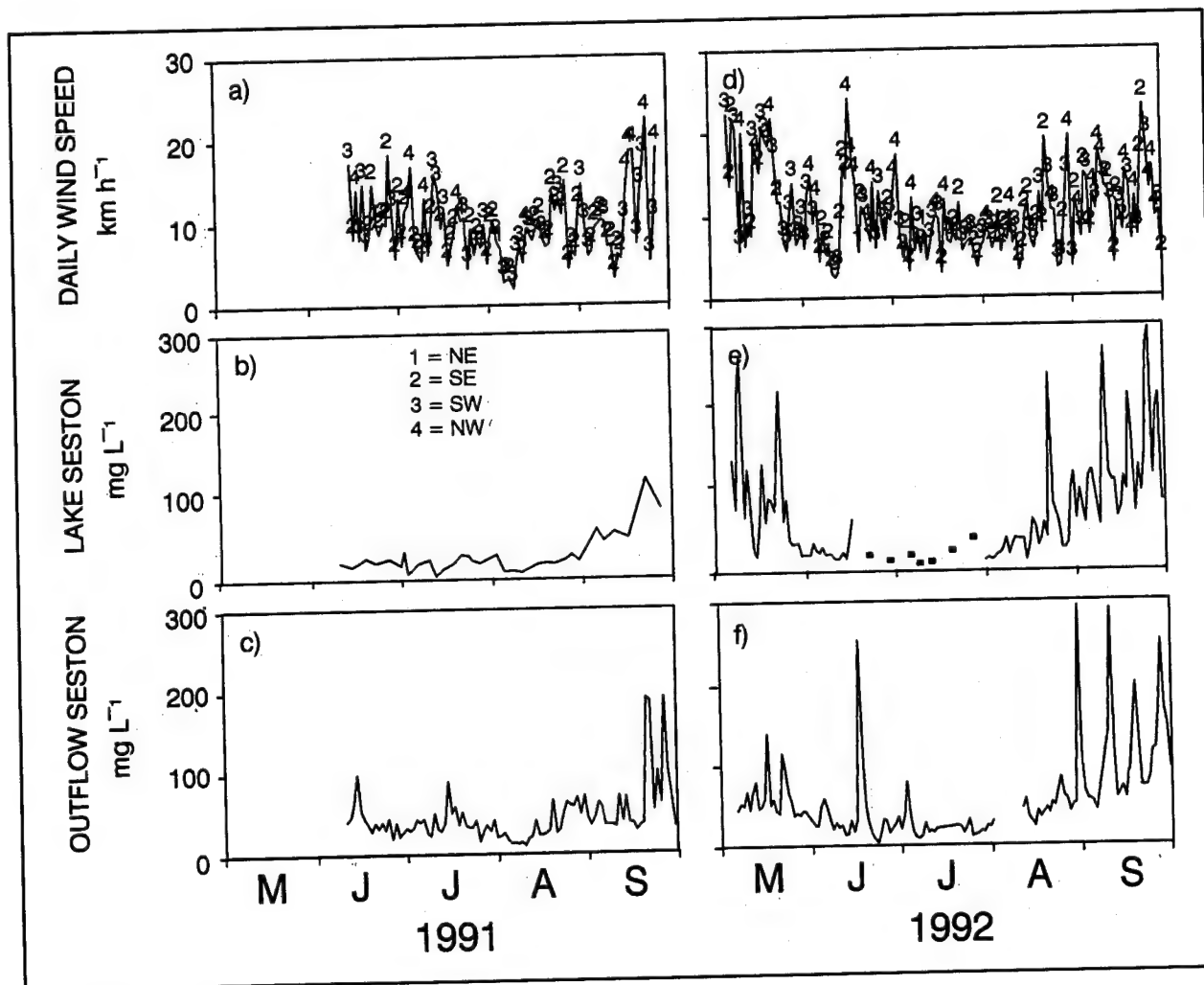


Figure 2. Variations in mean daily wind velocity (a and d), and seston concentrations in Marsh Lake (b and e) and the outflow (c and f) during 1991 and 1992. Seston concentrations in Marsh Lake represent the average in concentration from sta 1 and 2

markedly in late September (i.e., 100 mg L^{-1}) in association with high mean daily wind velocities of 19 km h^{-1} . A similar seasonal pattern was observed for seston concentrations in the outflow of Marsh Lake during 1991 (Figure 2c), suggesting the discharge of resuspended sediment.

During 1992, mean daily wind velocities were generally greatest in May and late August through September, and much lower during late-June through mid-August (Figure 2d). However, the highest mean daily wind velocity of 24 km h^{-1} occurred in mid-June of that year. Seston concentrations increased substantially in both Marsh Lake and the outflow

(i.e., $100\text{--}300 \text{ mg L}^{-1}$) during these windy periods in 1992 (Figures 2e and 2f). In contrast, seston concentrations ($>50 \text{ mg L}^{-1}$) were much lower during the calmer summer months.

Although we predicted from the wave model (Carper and Bachmann 1984) that sediment resuspension would occur at wind velocities of only 10.5 to 15 km h^{-1} , depending on wind direction (Table 2), the actual critical wind velocity for sediment resuspension was about 20 km h^{-1} in 1991, based on relationships between mean daily wind speed and Marsh Lake seston concentrations (Figure 3a). At these wind velocities, which blew primarily from the northwest direction, seston concentrations

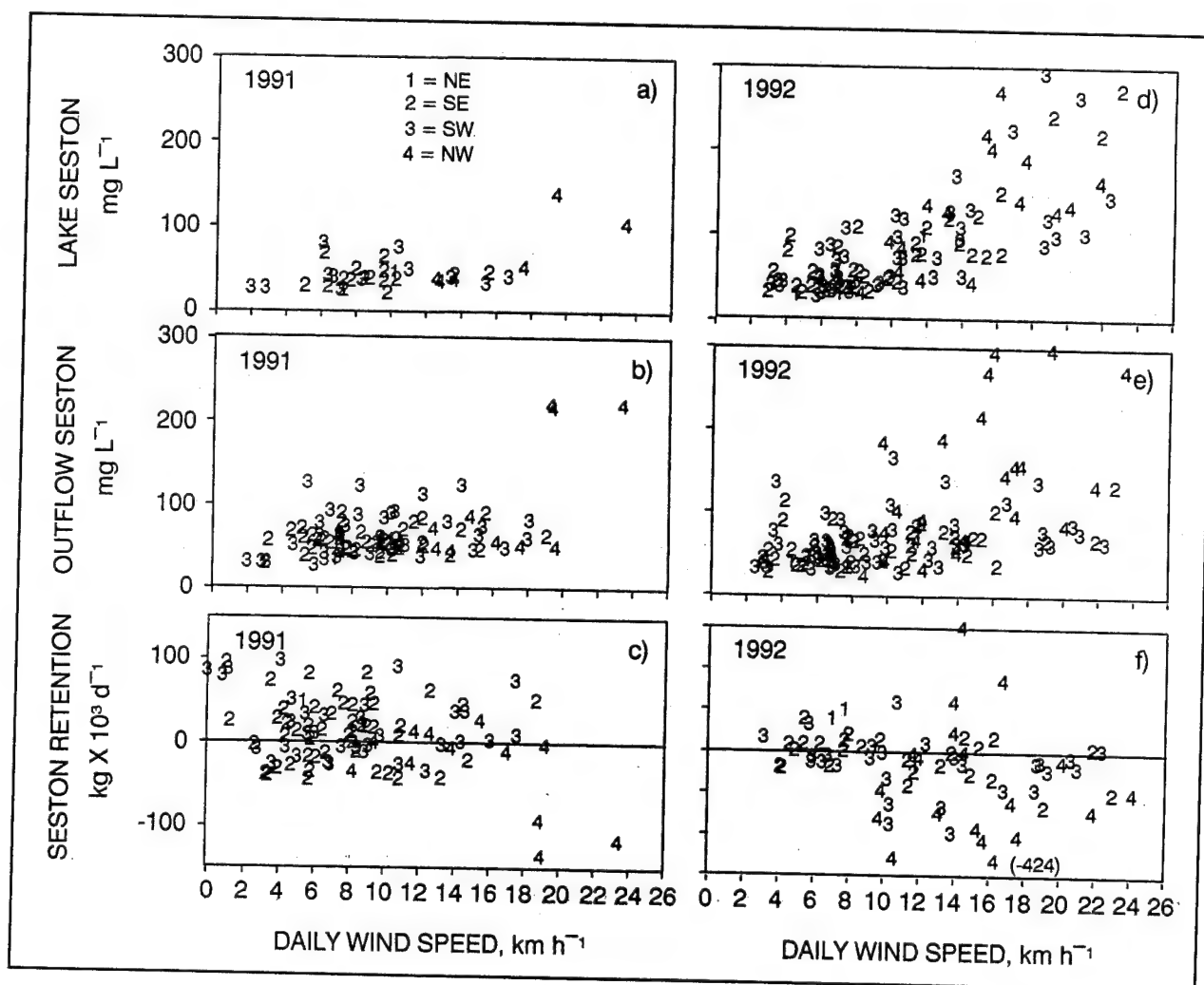


Figure 3. Relationships between mean daily wind velocity and seston concentrations in Marsh Lake (a and d) and the outflow (b and e), and daily retention of seston in the lake (c and f) during 1991 and 1992. Seston concentrations in Marsh Lake represent the average in concentration from sta 1 and 2

were also elevated in the outflow in 1991 (Figure 3b), and the seston retention rate was high and extremely negative (i.e., $< -100 \times 10^3$ kg/d; Figure 3c), indicating a net loss of sediment from the system during sediment resuspension events. The discrepancy between predicted and observed critical wind velocities may be attributed to the presence of high densities of submersed aquatic macrophytes which potentially dampened wave activity and sediment resuspension in 1991.

In contrast, relationships between mean daily wind speed and seston concentrations in Marsh Lake suggested that a critical wind velocity of about 12 km h^{-1} was required for sediment resuspension in 1992 (Figure 3d). The decrease in the critical wind velocity in 1992 was within the range of predicted values determined from the wave model (Table 2), and coincided with an almost complete absence of submersed aquatic macrophytes in Marsh Lake during that year. Above the critical wind velocity of 12 km h^{-1} , seston concentrations increased in a linear fashion with increasing wind velocity ($p < 0.05$; $r^2 = 0.58$), regardless of wind direction. Below this critical wind velocity, seston concentrations were usually $< 50 \text{ mg L}^{-1}$. Anomalously high seston concentrations (i.e., between 50 and 100 mg L^{-1}) below this critical wind velocity were representative of seston samples collected one day after the occurrence of a sediment resuspension event (wind velocities $> 15 \text{ km h}^{-1}$). This pattern in 1992 suggested a time lag of at least 1 day for the recovery of seston concentrations to nominal values following wind-generated sediment resuspension.

Discussion

The presence or absence of aquatic vegetation appears to play an important role in sediment resuspension dynamics in Marsh Lake. As an apparent result of the presence of aquatic vegetation in 1991, the critical wind velocity required to cause sediment resuspension was 5 to nearly 10 km h^{-1} higher than the values predicted from the wave model. In contrast, the critical wind velocity was much lower in 1992, owing to the absence of

aquatic macrophytes. This change in the critical wind velocity from about 20 km h^{-1} in 1991 to 12 km h^{-1} in 1992 resulted in a > 25 percent increase in the frequency of occurrence of sediment resuspension in 1992 (i.e., from 5 percent occurrence in 1991 to 32 percent occurrence in 1992). The wave model predicted a 30 percent frequency of occurrence in sediment resuspension during both years, based on the wind velocity record, which was similar to the observed frequency of occurrence in 1992 when submersed macrophytes were virtually absent in the lake. These results strongly suggest that aquatic vegetation was beneficial in reducing the occurrence of sediment resuspension in Marsh Lake by dampening wave activity.

Several studies have shown that emergent and submersed macrophytes can reduce and/or redirect currents and wave activity that promote sediment resuspension (Fonseca et al., 1982; Gregg and Rose 1982; Madson and Warncke 1983; Eckman, Duggins, and Sewell 1989). Others have shown that sediment resuspension and erosion are reduced, while net sedimentation is enhanced, in shallow regions of lakes occupied by aquatic vegetation (Moeller and Wetzel 1988; Deiter 1990; James and Barko 1990; Anderson 1990; Petticrew and Kalff 1991, 1992). In particular, although nearly 80 percent of the sediment surface area of shallow Lake Tämnen (mean depth = 1.5 m) is potentially resuspended as a result of wind and wave activity, all vegetated regions along the shoreline represent zones where sediments accumulate (Hellström 1991).

Sediment transport from Marsh Lake during periods of resuspension could have an important impact on Lac Qui Parle Reservoir, located immediately downstream, by both exacerbating accretion rates and accelerating the reduction in water storage capacity. In addition, accumulation of POM associated with resuspended sediment from Marsh Lake could have an impact on dissolved oxygen conditions in Lac Qui Parle Reservoir during ice formation. For instance, Gunnison and Barko (1988) and James et al. (1992) suggested that the resuspension and downstream transport of

sediment associated chemical oxygen-demanding materials was responsible for the rapid development of anoxia during winter drawdown in Big Eau Pleine Reservoir, WI. Discharge of resuspended phosphorus and other nutrients could also accelerate eutrophication and algal productivity in impoundments located downstream of Marsh Lake.

The results of these studies have implications for management alternatives to improve habitat and water quality in both Marsh Lake and Lac Qui Parle Reservoir. Techniques need to be developed to maintain the abundance of aquatic macrophytes in Marsh Lake to reduce sediment resuspension. These techniques might include periodic drawdown for sediment consolidation and seed germination, macrophyte transplanting, regulating pool elevation at least initially for the establishment of both submersed and emergent vegetation, and construction of artificial islands in strategic locations to reduce wind fetch and turbidity associated with sediment resuspension. Application of these techniques would encourage the development and maintenance of extensive stands of aquatic vegetation, thereby creating valuable habitat for waterfowl and limiting wind-driven sediment resuspension and sediment discharge to Lac Qui Parle Reservoir.

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Water Movement in Submersed Macrophyte Beds

by

Craig S. Smith¹ and William F. James¹

This report summarizes studies of water movement in submersed macrophyte beds, conducted in Eau Galle Reservoir, WI, and Gunterville Reservoir, AL. Water movement mediates the exchange of dissolved and suspended materials, including nutrients, dissolved oxygen, and suspended sediments. These exchanges have potentially important implications for both vegetated and open water areas, particularly since they occur between areas dominated by different ecological life forms and processes. The littoral zone can be an important source of phosphorus (P) and other nutrients to the pelagic zone of lakes and reservoirs. Rooted submersed macrophytes mobilize sediment P by root uptake and senescence (Barko and Smart 1980; Carpenter 1980; Smith and Adams 1986). Macrophytes can also influence the release of nutrients from sediments, by influencing pH and other environmental variables. Water circulation carries P and other nutrients mobilized by either of these mechanisms from vegetated regions to nearby open water areas. By altering water movements, macrophytes also influence sedimentation (James and Barko 1990; Petticrew and Kalff 1991), which in turn affects nutrient availability in the sediments. Typically, sedimentation rates are reduced within the center of macrophyte beds (Eakin and Barko 1989), although they can be elevated on the edges.

Water circulation through plant beds is also a primary determinant of the contact time of herbicides applied to control submersed macrophytes. Since successful control of plants by herbicides depends on maintaining adequate concentrations for sufficient time periods, rates of water circulation determine which herbicides can be used, at what rates they need to be applied, and whether or not they will successfully provide control.

Our studies focused on water movements driven by convection and by water level fluctuations resulting from changes in reservoir discharge. Even in the absence of wind, these mechanisms can produce relatively short water residence times in the littoral zone. Convective circulation is driven by horizontal water temperature (and density) gradients that develop when adjacent shallow and deep regions with differing surface-to-volume ratios are heated or cooled (Figure 1). During daytime heating, water in shallow regions heats more rapidly than in deeper regions (i.e., differential heating), resulting in horizontal movement of shallow water as a surface flow over cooler water in the deep region (Monismith, Imberger, and Morison 1990). During nighttime cooling, the opposite pattern occurs. Water in shallow regions cools more rapidly than in deeper regions (i.e., differential cooling), resulting in horizontal movement of shallow water as an underflow below warmer water in the deep region, until a similar density stratum is encountered in the water column (Monismith, Imberger, and Morison 1990).

Eau Galle Reservoir

In Eau Galle Reservoir we examined the potential for convective water exchanges in two small, sheltered embayments. During 1988, 1989, 1991, and 1993 we established transects of recording stations and recorded temperature measurements at closely spaced depth intervals. On eight occasions during 1988 and 1989, rhodamine WT dye was injected to trace water movements. Phosphorus (P) concentrations were measured along the temperature transect during 1988 and 1989. In 1991 and 1993, temperature recording

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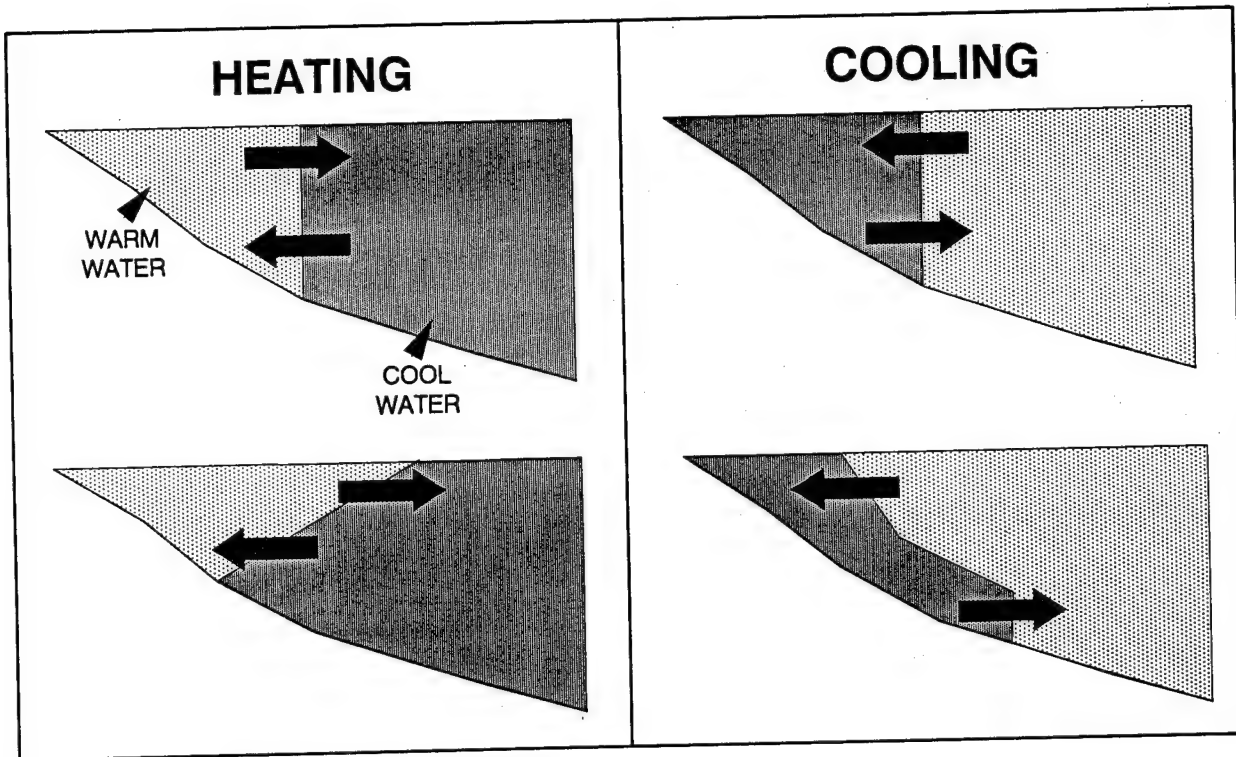


Figure 1. Differential heating and cooling produce horizontal temperature gradients that lead to convective circulation

stations were established at very close spatial intervals and temperature and meteorological measurements were made more frequently (every 5 minutes).

Differential cooling occurred frequently at night in the embayments of Eau Galle reservoir. In 1989, temperature patterns conducive to convective circulation developed on 31 to 95 percent of summer nights, depending on the month.

Injected dye invariably moved in a pattern characteristic of cooling-driven convective circulation (Figure 2). Phosphorus concentration data indicated that convective water movements were transporting significant quantities of P from the littoral zone into the pelagic zone (James and Barko 1991b).

Close-spaced, high-frequency temperature data from August 1991 were used to estimate mean areal convective exchange rates during periods of differential cooling in the southwest bay, using the heat budget model of Stefan, Horsch, and Barko (1989). Details of this effort are described in James and Barko (1994).

Differential cooling occurred on every night of the study. The overall mean areal convective rate calculated using the model ($0.00066 \text{ m}^3/\text{m s}$) was comparable to the mean convective exchange rate estimated for the summer of 1989 (June - August) based on rhodamine WT dye movement ($0.0005 \text{ m}^3/\text{m s}$) (James and Barko 1991b). The similarity in rates suggests that the heat budget model provides a good estimate of convective exchange rates for this reservoir.

Guntersville Reservoir

Studies of temperature patterns and water movement were conducted in two embayments within the Guntersville Reservoir in northeastern Alabama (Figure 3). The Minky Creek embayment is a large, open embayment located at the lower end of the reservoir near the dam. The Mud Creek embayment is located in a drowned tributary near the center of the reservoir. Though large overall, the Mud Creek embayment is segmented into smaller areas by several constrictions and is isolated from the Tennessee River by the natural levees of

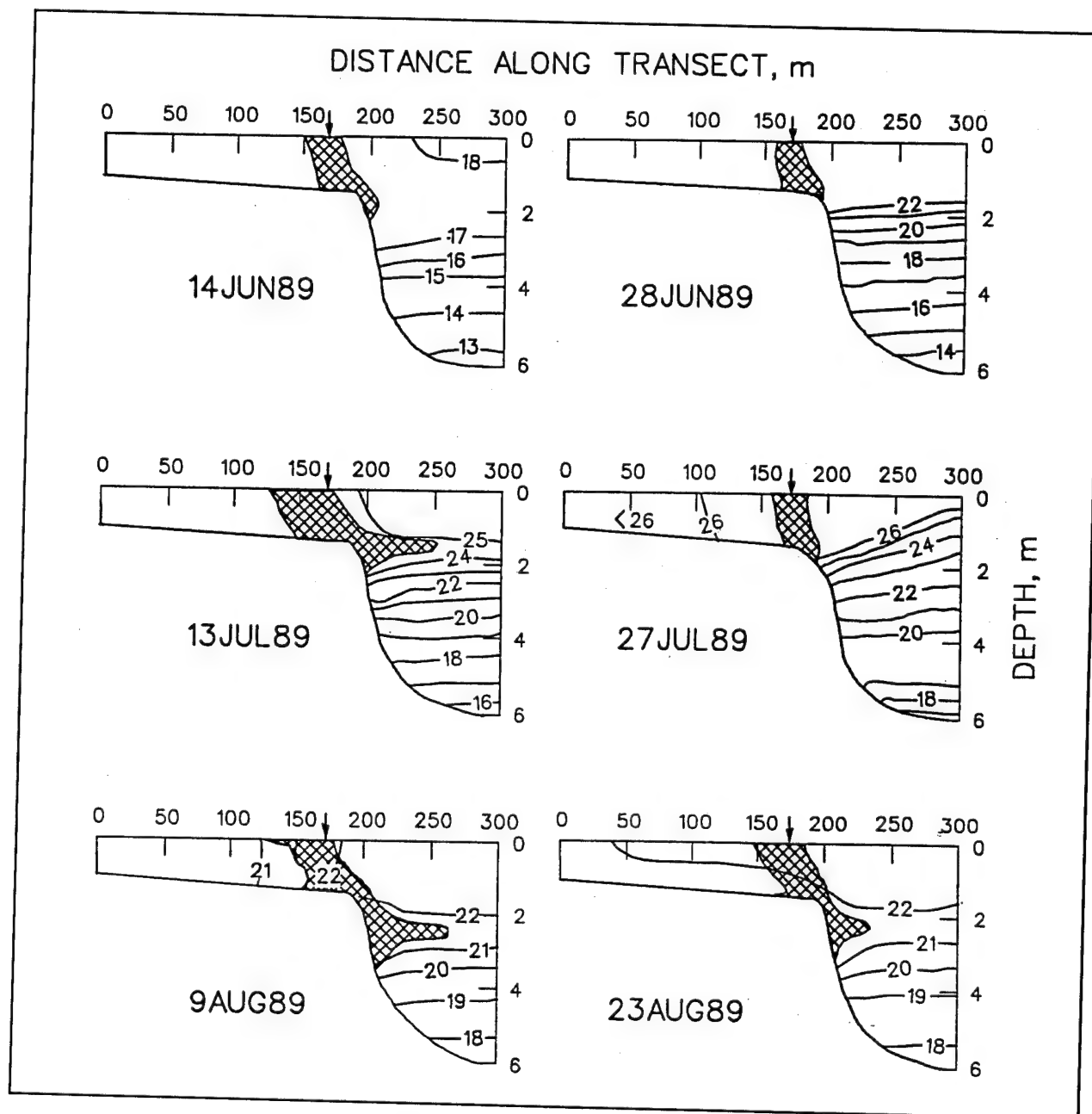


Figure 2. Temperature patterns and dye distribution in the northwest bay of Eau Galle Reservoir on six mornings in 1989

the river, to which it connects via a narrow, winding channel.

Minky Creek

We used vertical and horizontal variations in water temperature to examine differential heating and cooling in the Minky Creek embayment during 1990, 1991, and 1992. Our objectives were to identify temperature patterns likely to produce convective circulation

in this embayment and to document the frequency of their occurrence. The embayment was densely vegetated by *Myriophyllum spicatum* in 1988, but submersed plants in the embayment declined prior to 1990, leaving the site unvegetated during our study.

Even in this relatively large, exposed embayment, heating and cooling were often sufficient to greatly alter vertical and horizontal temperature gradients. On sunny days, shallow

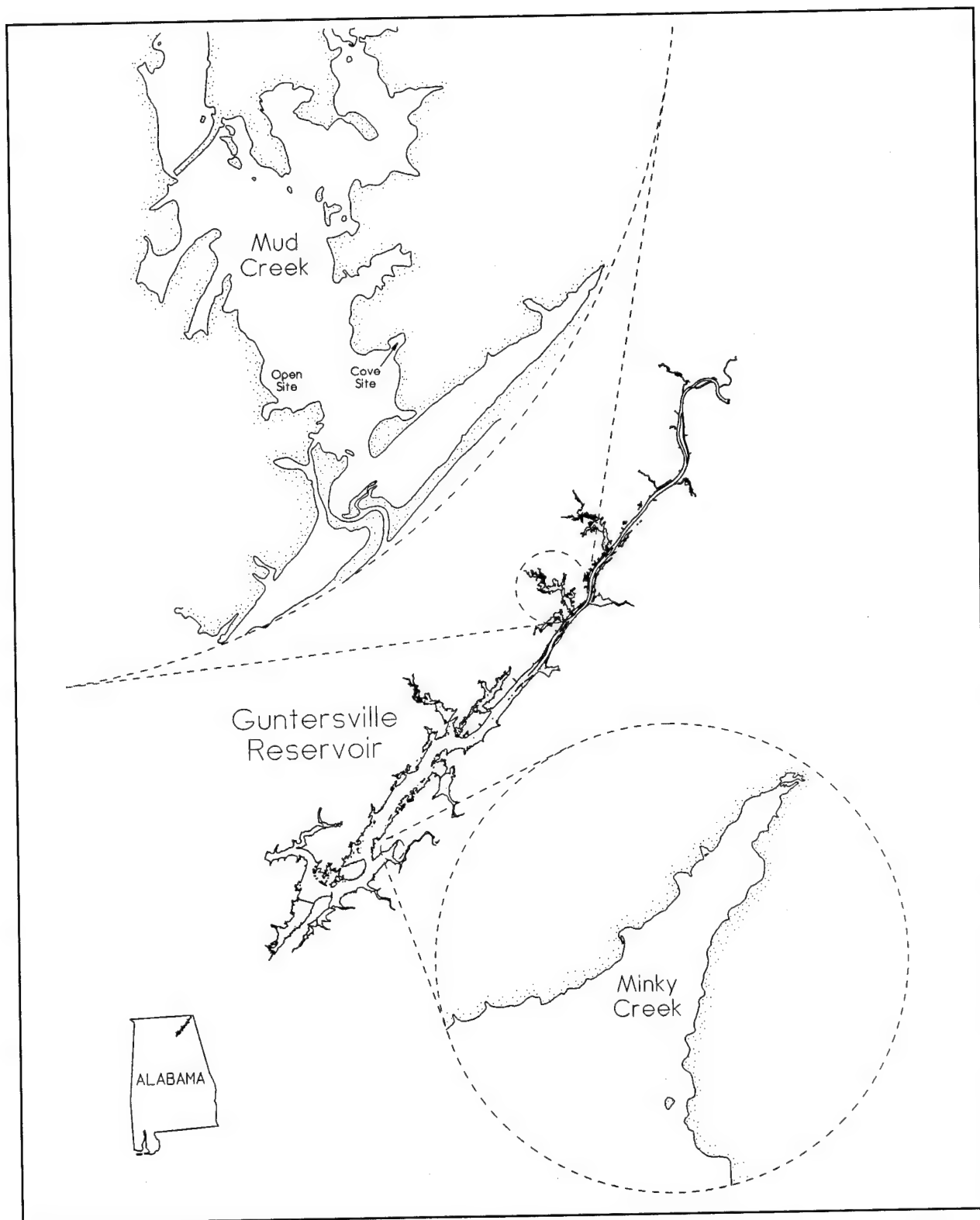


Figure 3. Location of the Minky Creek and Mud Creek embayments in Guntersville Reservoir

areas often became much warmer than even the surface layers at deeper locations. On calm days, temperature gradients of $1\text{ }^{\circ}\text{C}/\text{m}$ or greater developed within 0.25 m of the surface. During periods of nighttime cooling, temperature patterns developed that suggest the existence of convectively driven underflows of cooler water.

The results of a dye tracer experiment conducted October 16, 1991, confirmed that, at least on this date, cooler water was moving outward along the bottom of the embayment. Temperatures measured during the night of October 15 and the morning of October 16 (Figure 4) suggest considerable nighttime cooling with an apparent underflow of cooler water at the central stations from 0800 through 1200 hr. Movement of rhodamine WT dye was tracked following a surface-to-bottom injection made between 0800 and 0820 hr along the central transect (Figure 5). The dye cloud moved toward shore in near-surface layers (from approximately 0 to 1.25 m) and away from shore in deeper layers (1.5 m and deeper). By the end of the study, a portion of the surface dye cloud had been deflected by wind around the periphery of the embayment and back across the central transect, where it appeared as a separate dye cloud near the surface at station 6. This illustrates the complex, three-dimensional circulation patterns produced by the combined effects of convection and wind.

Mud Creek

In 1993, studies of hydraulic circulation were moved upstream to the Mud Creek embayment. Submersed vegetation in the Minky Creek embayment had not recovered by 1993, and the lack of submersed vegetation in the embayment made it impossible to examine the effects of vegetation on water circulation or to determine the rates of water movement through vegetated areas. The Mud Creek location provided an opportunity to examine water circulation in more sheltered, vegetated locations. Two sites varying in exposure were selected, one in a small cove and the other located along an open section of shoreline (Figure 6). Both sites contained dense beds of *Myriophyllum spicatum*. From May

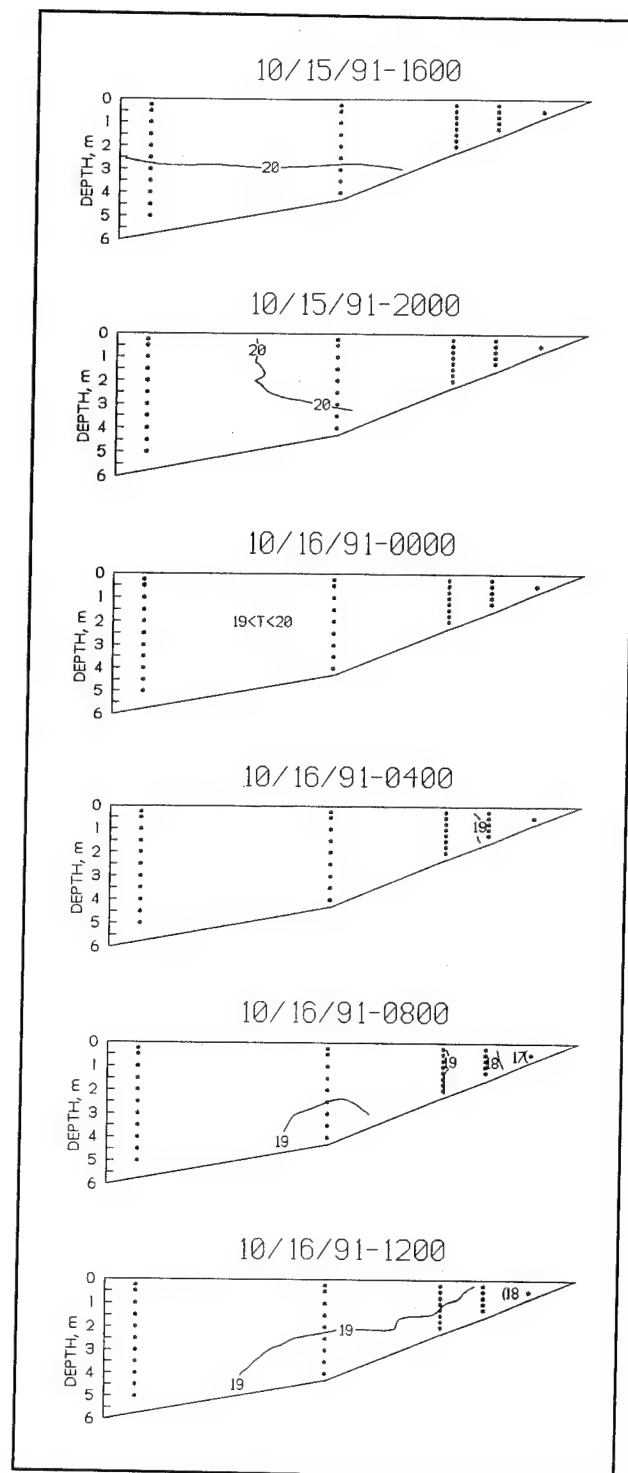


Figure 4. Temperature patterns in the Minky Creek embayment on October 15 and 16, 1991, when substantial differential cooling was occurring

through November of 1993 water temperature and meteorological conditions were recorded and many dye injection studies were conducted in the two Mud Creek sites.

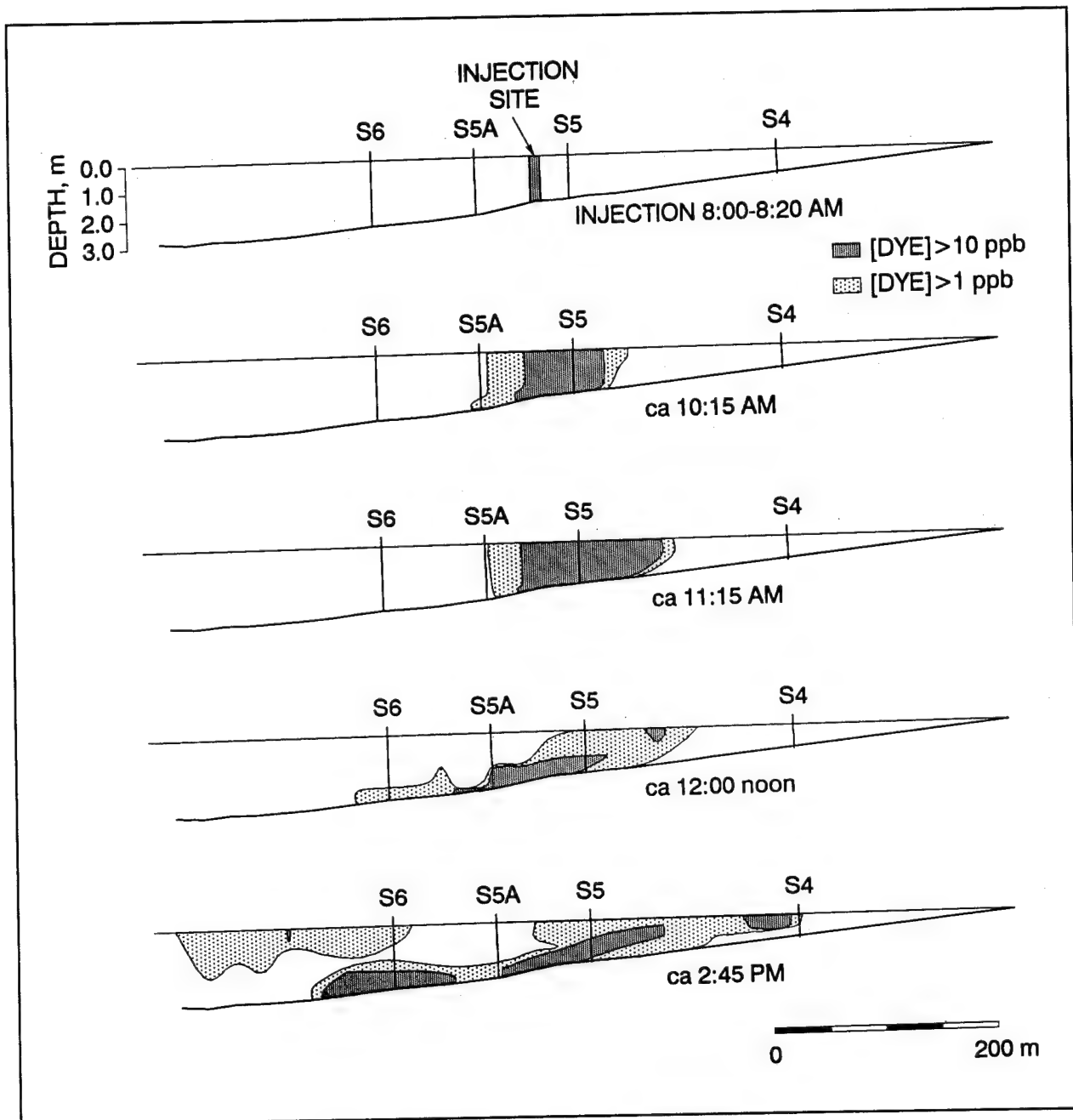


Figure 5. Movement of rhodamine WT dye injected into the Minky Creek embayment on October 16, 1991

In the cove site, dye moved primarily along the transect axis, so analysis of water movement in the cove concentrates on movement along this axis. On all dates examined, patterns of water movement were relatively consistent (Figure 7). Prior to 1100 hr, water on the surface and bottom of the cove moved toward the back of the cove, usually at a moderate rate. After approximately 1100 hr, bottom water moved rapidly out toward the

mouth of the cove. Surface water moved relatively slowly after 1100 hr, and the direction of movement varied. Around 1800 hr, surface water movement began moving toward the mouth of the cove.

There was a strong correspondence between water level changes and the direction and rate of water movement in the cove. At the Mud Creek site, the operation of Nickajack Dam

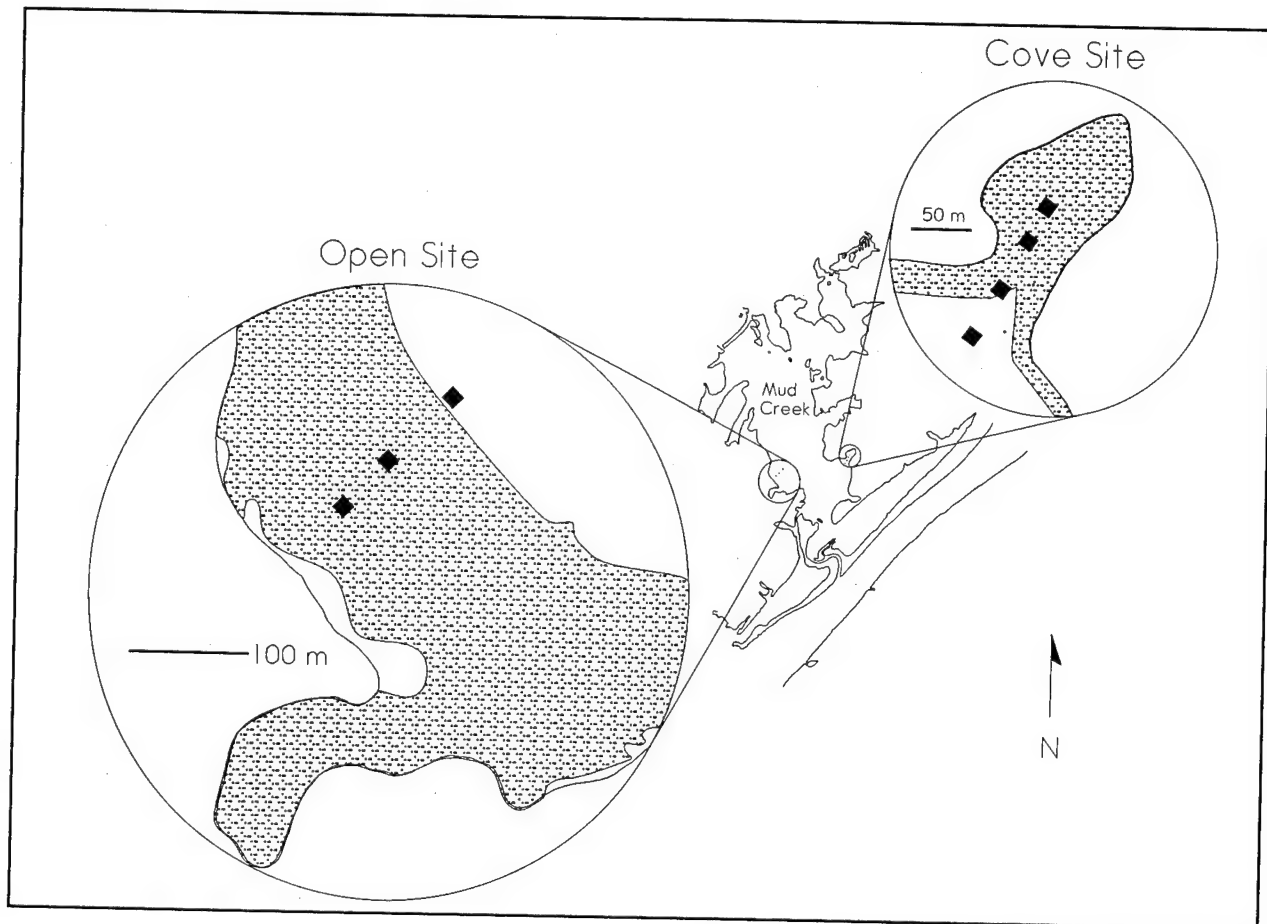


Figure 6. Location of the cove and open sites in the Mud Creek embayment. Shading indicates the extent of macrophyte beds. Black squares show the locations of temperature and/or meteorological monitoring stations

produced a cycle of rising water levels from approximately 0600 hr until around 1000 hr or 1100 hr and falling water levels from then until the following morning. As water levels rose, both surface and bottom water moved toward the rear of the cove. After the peak, as water levels began to fall, water on the bottom switched directions almost immediately and began to move rapidly outward. Surface water movement slowed as water levels began to recede, but surface water did not begin to move outward until water levels had been dropping for several hours.

Water movement rates and patterns in the open site were highly variable. Surface and bottom water masses typically moved in different directions. The direction of water movement was not at all consistent from study to study and often changed during the course of a single study. No relationships

with wind or convection were evident. In this location, most of the studies were conducted during periods of little water level change and effects of reservoir operation were not usually discernible. One exception to this generalization occurred on October 7, 1993, when water levels rose 5 cm during the first 2.5 hr of the study and then fell 4 cm during the last 3 hr. On this date, surface and bottom water masses moved in a pattern similar to that observed in the cove site (Figure 8). As the water level rose, both surface and bottom water masses moved in a direction that was approximately upstream. As the water level began to fall, movement on the surface continued in the same direction as before but the bottom water mass changed direction and began to move back toward the mouth of the creek.

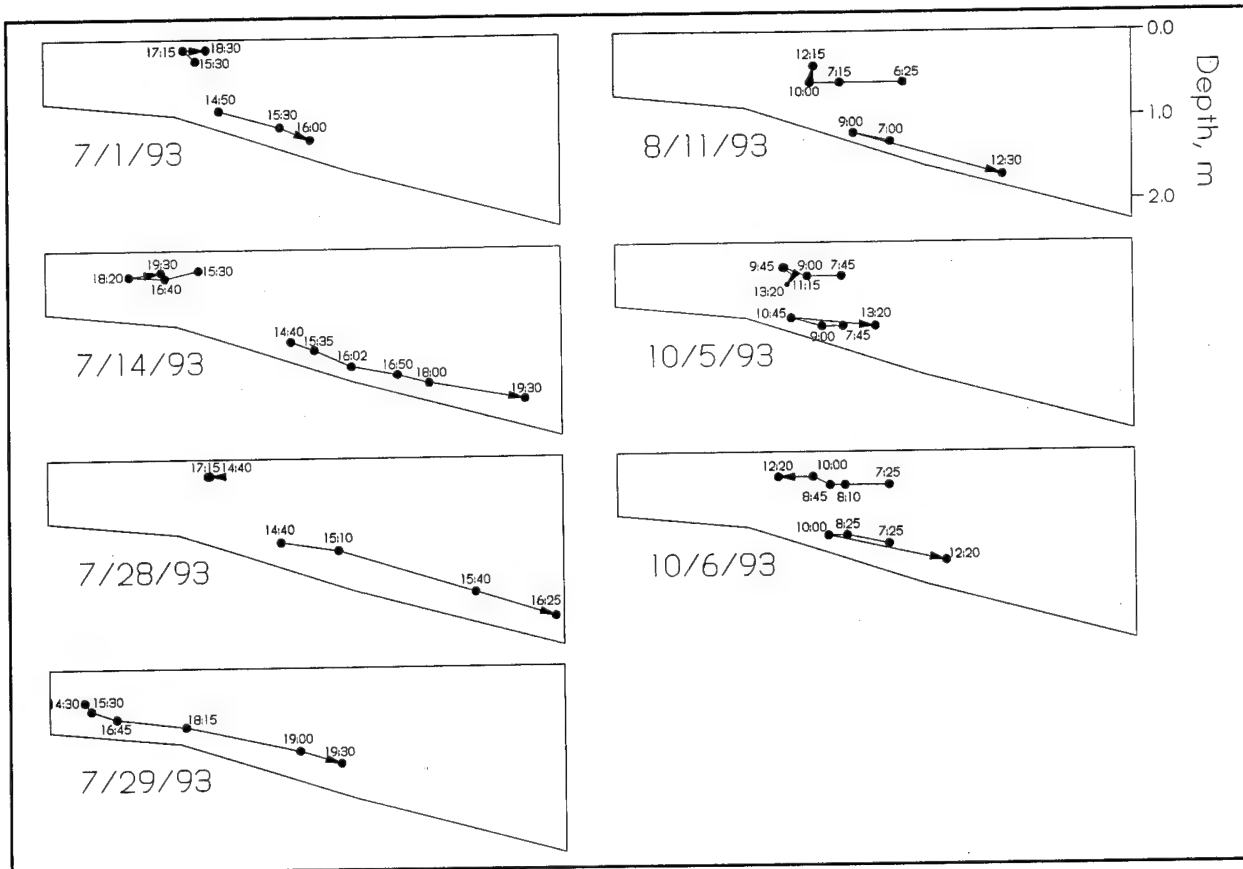


Figure 7. Movement of the centers of mass of shallow (<1.0 m) and deep (≥ 1.0 m) dye clouds in the Mud Creek cove site

Temperature patterns in the cove reflected the retention of surface water in the back of the cove while bottom water drained outward, as shown in temperature patterns from July 1 (Figure 9). Heating during the day produced a very warm layer of water on the surface. Instead of flowing outward as it would in the case of convective circulation, warm surface water was retained in the cove as water levels fell. The result was a zone of warm temperatures on the surface that receded toward the back of the cove as the afternoon and evening progressed. The retention of warm water in the back of the cove led to very high temperatures. Surface temperatures in excess of 30°C were recorded on 41 days and temperatures in excess of 40°C were recorded on 2 days.

Discussion

Convection due to nighttime cooling was very important in embayments of Eau Galle

Reservoir. Relative to other mechanisms of water circulation, convection is probably more important in Eau Galle Reservoir than in the other sites we examined for several reasons. Eau Galle Reservoir is small and sheltered from wind by its position at the bottom of a steep valley. It is located in a region where nighttime cooling is frequently substantial. Thus, the reservoir is an ideal site for detecting convection-driven currents.

Effects of convection were less apparent in the two sites in Guntersville Reservoir. The Minky Creek site is a large, exposed embayment, and the effects of wind made it much more difficult to detect convective water circulation. Nonetheless, temperature patterns conducive to convection were relatively common, and the results of a single dye study showed that convection was important on that occasion. It is often difficult to separate wind-generated circulation patterns from those generated by differential heating and

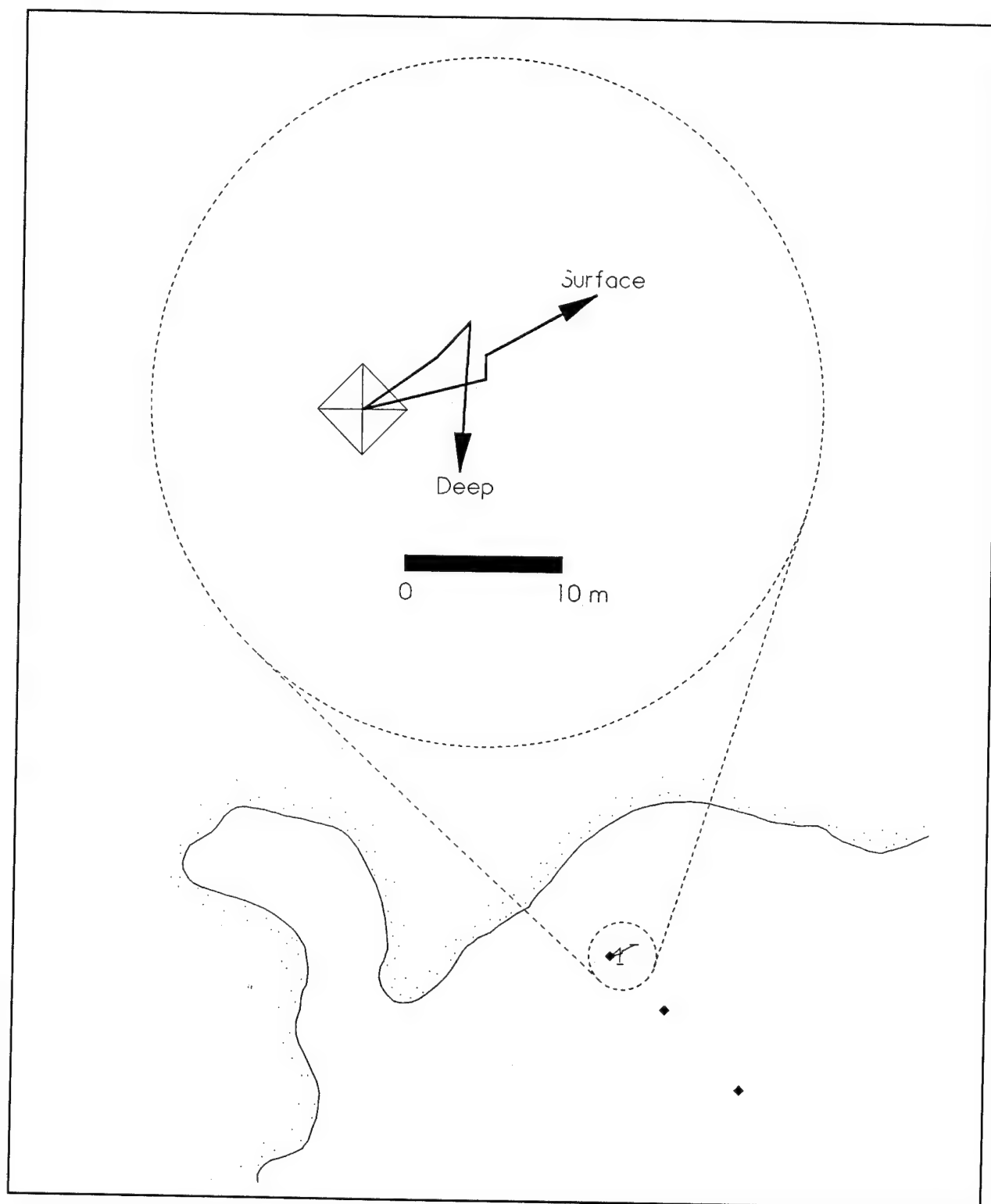


Figure 8. Movement of the centers of mass of shallow (<1.0 m) and deep (≥1.0 m) dye clouds during the dye injection study conducted October 7, 1993, in the Mud Creek open site

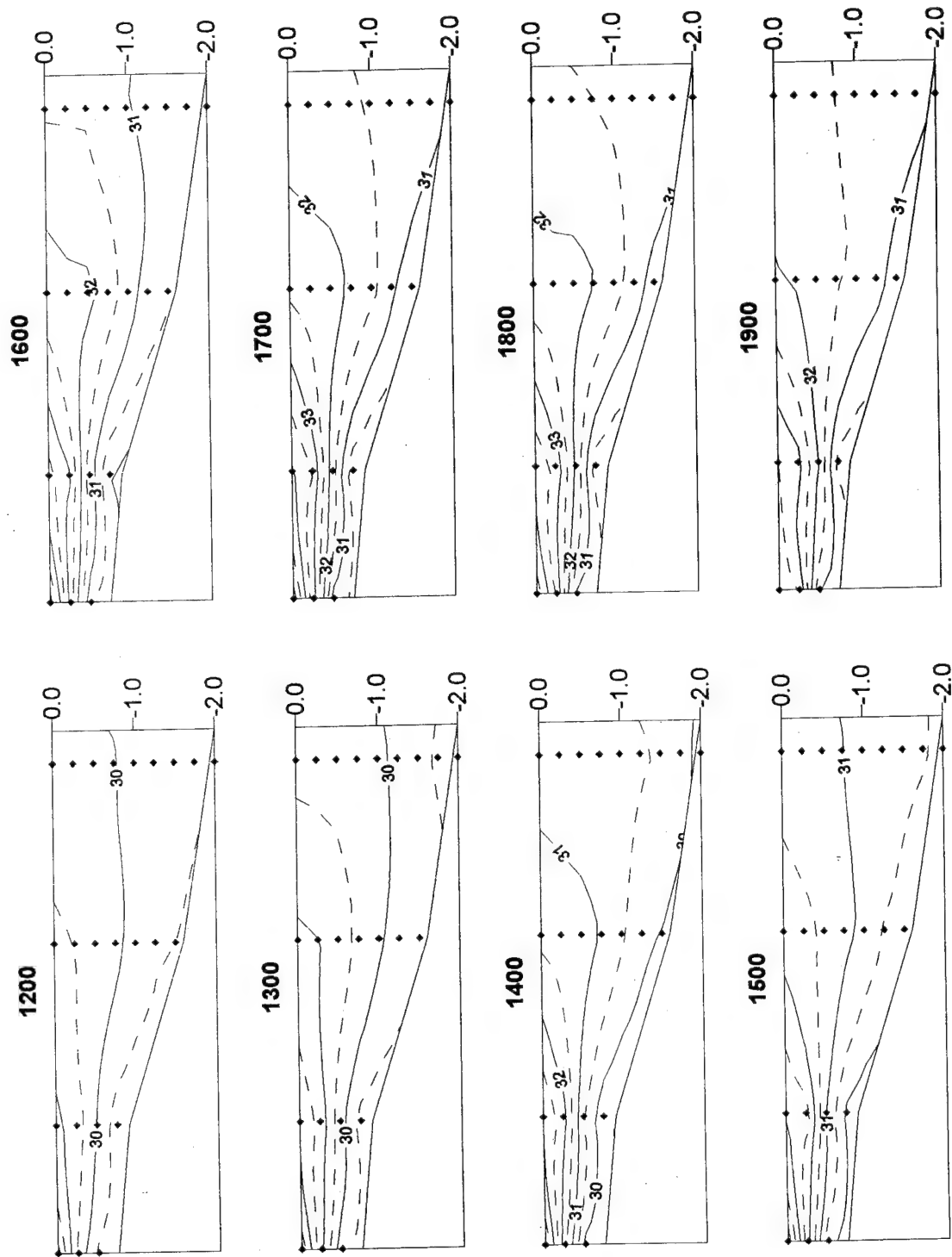


Figure 9. Water temperatures in the Mud Creek cove site, July 1, 1993

cooling on the basis of water temperature alone (Monismith, Imberger, and Morison 1990) and it is not clear how often convectively driven exchanges, like that observed during the dye study, actually occur.

Water circulation in the Mud Creek embayment was largely dominated by the effects of water level changes caused by the operation of Nickajack Dam. Effects of operation were particularly apparent in the cove site, where water circulation showed a predictable pattern coinciding with the daily cycle of water level changes. In this location rising water levels pushed both surface and bottom water back into the cove. As the water level fell, bottom water drained from the cove and warm, often very warm, surface water was retained in the back of the cove.

Effects of vegetation on water circulation were not readily apparent from results of our studies. Aquatic macrophytes both dampen wind effects and substantially reduce light penetration into the water column, resulting in the development of a very shallow (i.e., 25 cm or less) surface mixed layer. Shallow heat penetration prevents warming of the bottom waters in shallow regions, which may inhibit daytime convective circulation (Imberger and Patterson 1990). Since nighttime cooling is penetrative, it may be relatively unaffected in vegetation, or may be stronger because of reduced wind effects. Thus, circulation patterns may differ significantly between vegetated and unvegetated areas.

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Factors Contributing to the Decline of Eurasian Watermilfoil in TVA Reservoirs

by
Craig S. Smith¹

Upon invading a new location, populations of exotic submersed macrophytes often exhibit a characteristic pattern of expansion, dominance, and decline (Figure 1). Many factors have been proposed as possible causes of declines (Table 1), but little evidence supports the importance of any of these in causing specific declines (Carpenter 1980; Smith and Barko 1990).

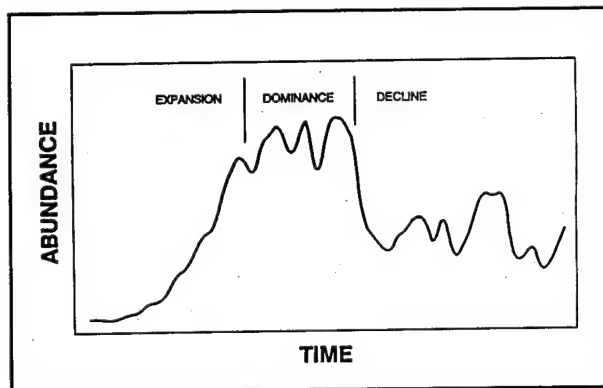


Figure 1. Typical population dynamics following invasion of a waterbody by an aggressive exotic aquatic plant such as Eurasian watermilfoil

Table 1
Possible Causes of Eurasian Watermilfoil Declines¹

Nutrient depletion
Toxin accumulation
Shading by phytoplankton or attached algae
Parasite(s) or pathogens(s)
Harvesting/Herbicides
Climatic fluctuations
Competition from other macrophytes
Insect herbivory (Painter and McCabe 1988)

¹ After Carpenter (1980) except as noted.

A substantial decline in submersed vegetation occurred throughout mainstream TVA reservoirs in 1989 through 1991 (Figure 2), coinciding with declines of submersed species in the upper Mississippi River (Rogers 1994) and the tidal Potomac River (Carter et al. 1993). During the summer of 1988 the eastern United States experienced a severe drought, and macrophyte coverage in the TVA reservoirs reached record levels. In the years 1989 through 1991 spring rainfall was above normal, resulting in high water flows and increased turbidity. Depending on the TVA reservoir, submersed macrophyte coverage declined over a 2- or 3-yr period. Improved growing conditions led to a partial recovery of aquatic vegetation by 1992 or 1993 in most of these reservoirs. One exception is Watts Bar Reservoir, the TVA reservoir into which Eurasian watermilfoil was first introduced, where recovery is not evident.

This paper describes studies of environmental factors associated with declines. Sites having populations of *Myriophyllum spicatum* (Eurasian watermilfoil) that were stable, declining/recovering, or nonexistent were examined. At each site, selected environmental conditions and plant cover were evaluated. Sediments were collected and returned to the laboratory, where they were bioassayed for their ability to support plant growth and their physical properties were analyzed.

Methods

Sites where *M. spicatum* populations had (1) remained constant, (2) declined, (3) declined and recovered, or (4) never invaded

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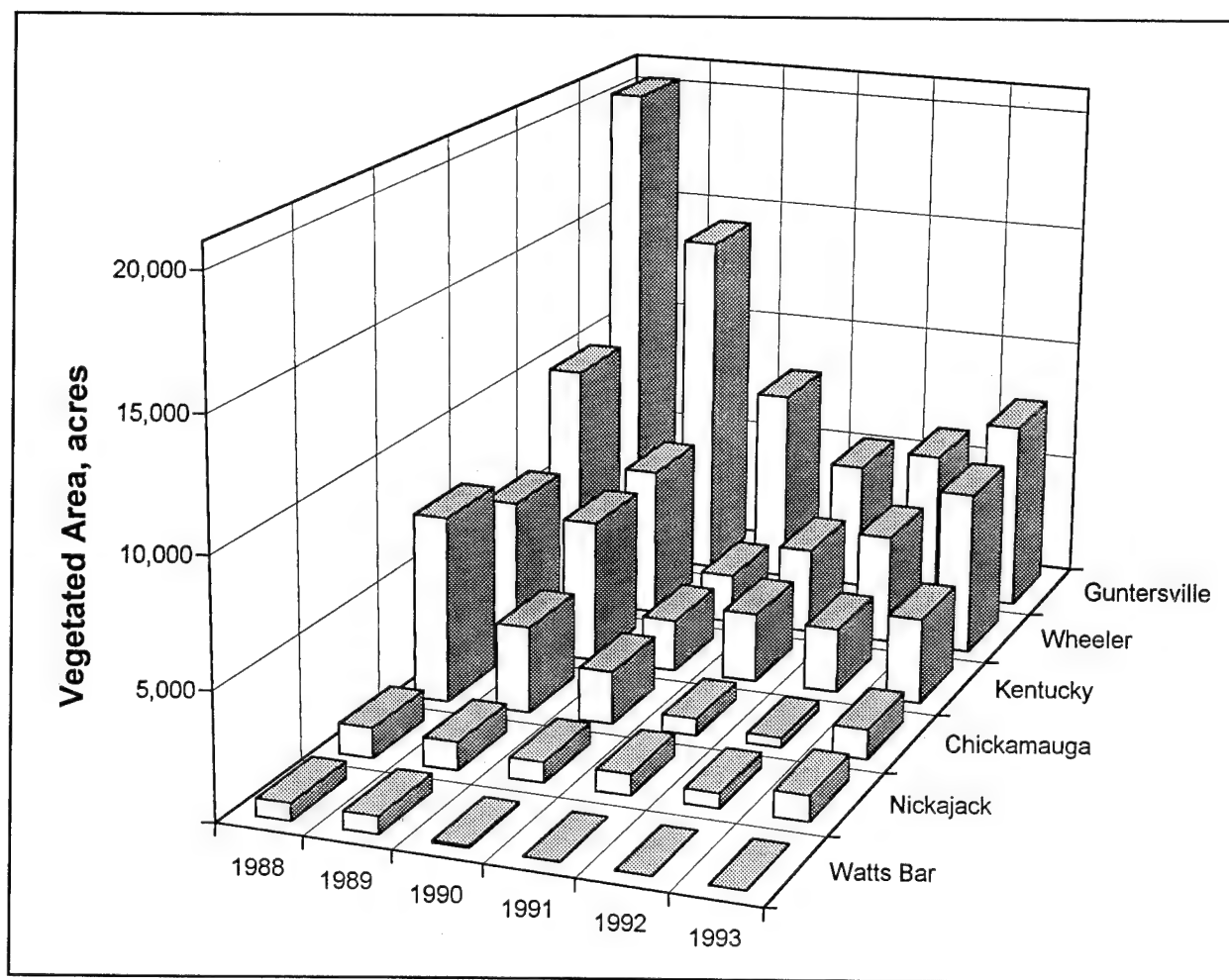


Figure 2. Macrophyte coverage in mainstream TVA reservoirs (from Bates and Smith 1994)

were examined to determine conditions that may have contributed to differences in the ability to support submersed macrophyte growth. Sites were located primarily in TVA reservoirs, where long-term records of plant cover are available from aerial photography, and in reservoirs on the adjacent Cumberland River, where *M. spicatum* has invaded more recently and populations are generally expanding (Figure 3).

The vegetation cover history of sites was determined from aerial photographs taken in 1987 or 1988, 1990, and 1992. Sites were selected to represent a range of environments (e.g., overbank, embayment, etc.). Sites were located where *M. spicatum* beds had been sufficiently large or were close enough to distinct landmarks so that it was possible to be reasonably sure of being within the former bed limits when collecting samples in the

field. Compass bearings to two to four recognizable landmarks were recorded at each sample location.

Basic environmental data were recorded for each sample site. These included a Secchi disk measurement, observations of plant cover, and information concerning any recent aquatic plant management. *Myriophyllum spicatum* cover was recorded in semi-quantitative categories ranging from absent to very dense. Presence and species composition of any other submersed vegetation were noted.

Sediments from the sample sites were collected and bioassayed for their ability to support *M. spicatum* growth. Sediment cores were collected in 10-cm-diameter PVC tubes 28 cm long. Core tubes were filled with intact cores of sediment by inserting the core tube vertically into the sediments, except

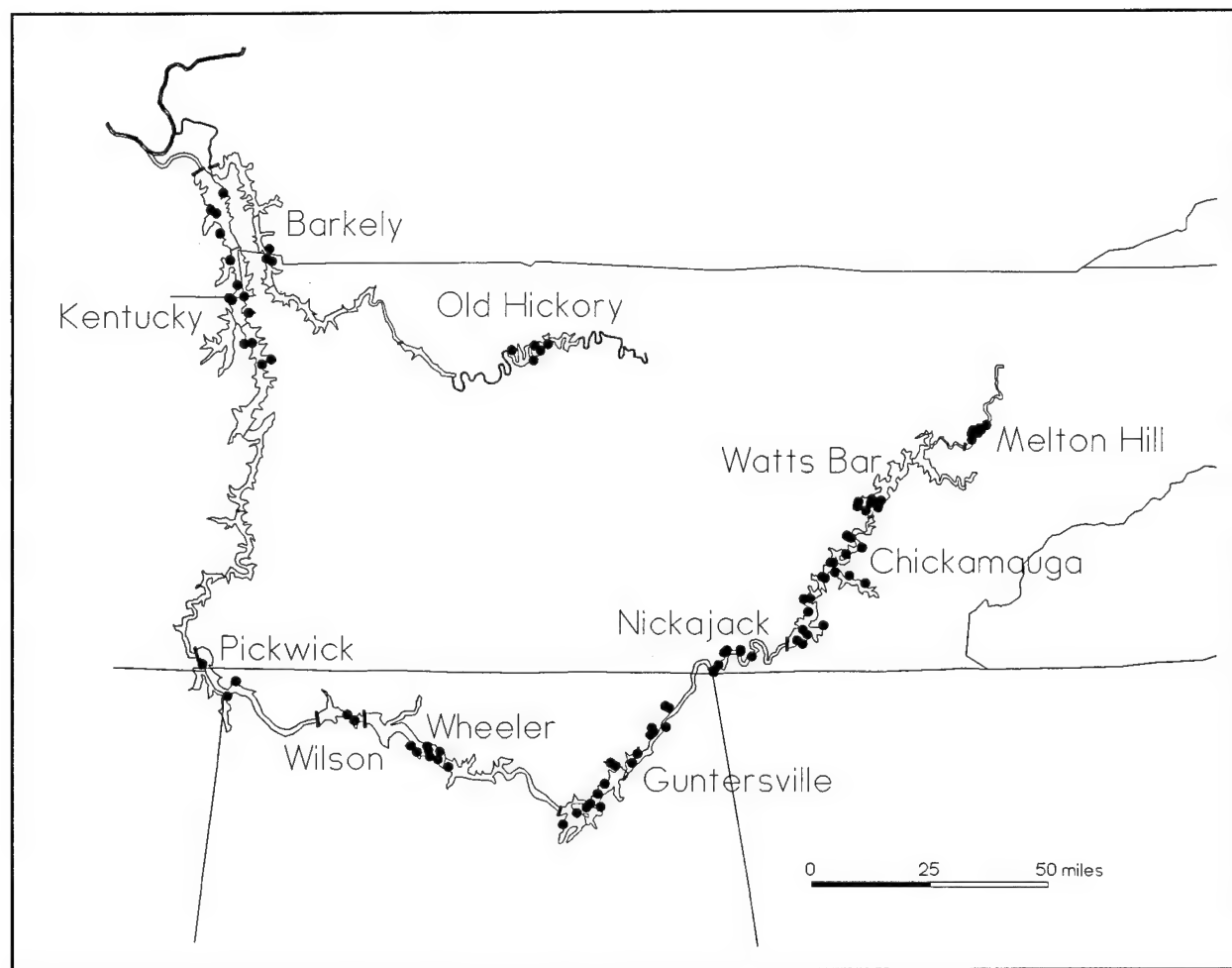


Figure 3. Locations where environmental data and sediment cores for bioassays were collected

where dense or rocky sediments made it impossible to insert the corer completely. In these cases additional surficial sediments were added to the top of the core to fill the tube. Cores were stored underwater at 5 °C until they could be bioassayed. Three experimental runs were required to bioassay all of the core samples.

Coarse sand was added to the top of the cores to minimize sediment resuspension and nutrient loss, and each core was planted with four 15-cm sprigs of *M. spicatum* from greenhouse stock cultures. For each experimental run, four additional core tubes were filled with sediments from Browns Lake, at the WES, and planted to provide a measure of *M. spicatum* growth on a known reference sediment. After planting, each core was inserted into the base of an individual acrylic column, which was then filled with growth medium

(Smart and Barko 1985). Columns were placed in a growth room, where they were maintained at 25 ± 1 °C and illuminated to approximately $400 \mu\text{mol quanta m}^{-2} \text{sec}^{-1}$ of photosynthetically active radiation for 14 hours/day. After 6 weeks of growth, aboveground parts of plants were harvested, dried to a constant weight at 80 °C, and weighed to determine the final biomass.

Sediments from the bioassays were analyzed to determine bulk density, organic content and particle size distribution. Sediment moisture content and bulk density were determined by weighing a known volume of wet sediment, drying the sample to a constant weight at 105 °C (Håkanson 1977), and reweighing it. Organic content was determined by loss on ignition at 550 °C (American Public Health Association 1985). Particle size distribution was determined by first sieving the

sediments through 6.3-mm and 2.0-mm mesh sieves and then determining the size distribution of finer particles by the hydrometer method of Patrick (1958).

Results

Results of the bioassays are shown in Figure 4. Sediments differed by approximately a factor of 5 in their ability to grow *M. spicatum*. Plants on 86 percent of the sediments produced biomass that was within the expected range for growth on the reference sediment, while the remaining sediments supported significantly less growth than the reference sediment. About one-half of the sediments supporting significantly less growth than the

reference sediment were from continuously vegetated sites; the remainder were from sites where vegetation had declined or had never been present.

None of the sediment parameters measured provided a particularly good predictor of biomass production. Biomass production was significantly correlated with the clay content of sediments, but this correlation only accounted for 22 percent of the variance ($r^2 = 0.22$).

Analysis of variance (ANOVA) detected no significant differences in biomass production on the sediments from areas having different *M. spicatum* histories (Figure 5). Similar analyses for the other parameters measured in this study revealed that only light

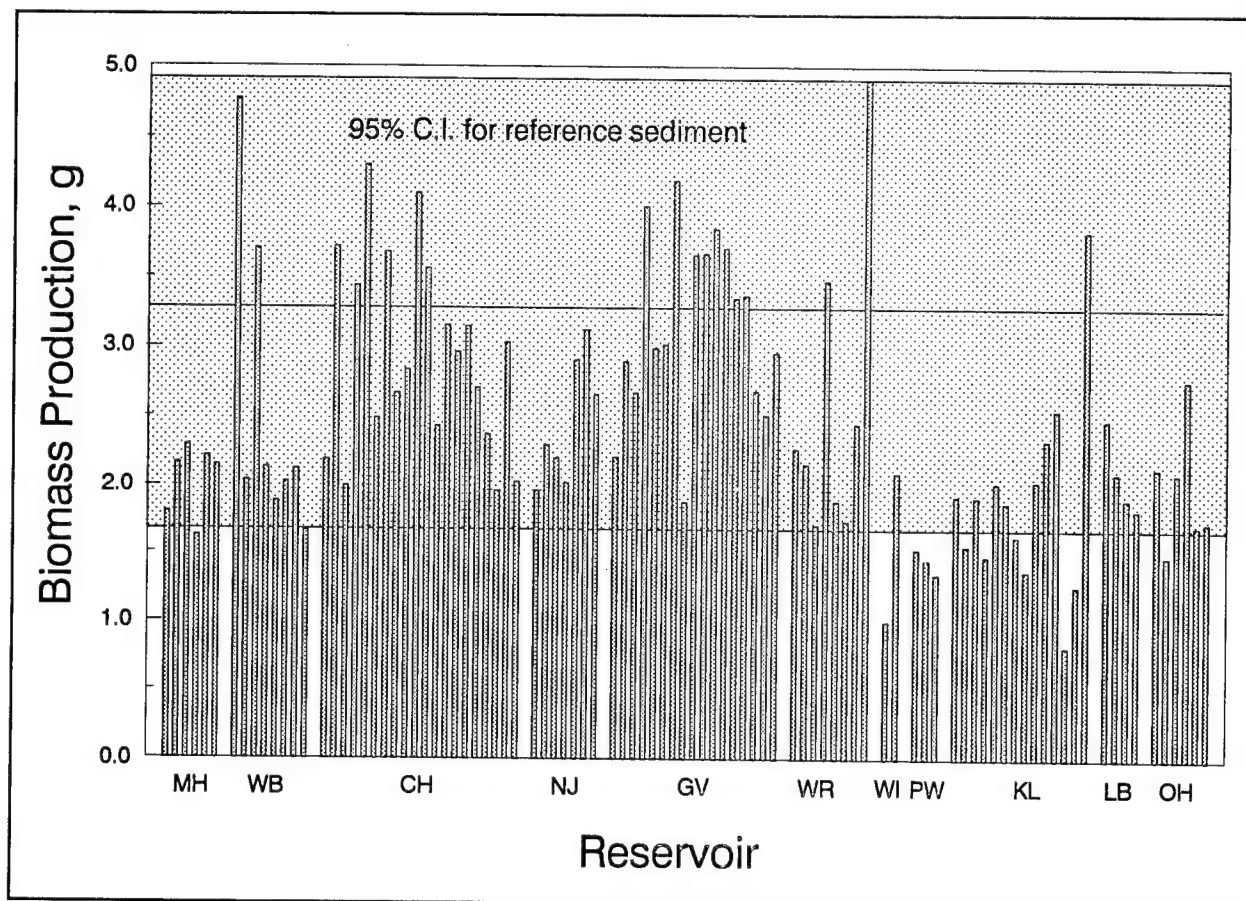


Figure 4. Growth of *M. spicatum* plants on sediments from Tennessee and Cumberland River reservoirs. Each bar represents the biomass produced on an individual sediment sample from the indicated reservoir (MH = Melton Hill, WB = Watts Bar, CH = Chickamauga, NJ = Nickajack, GV = Guntersville, WR = Wheeler, WI = Wilson, PW = Pickwick, KL = Kentucky Lake, LB = Lake Barkely, OH = Old Hickory). The shaded region indicates the 95 percent confidence zone for the growth of plants on reference sediment from Browns Lake

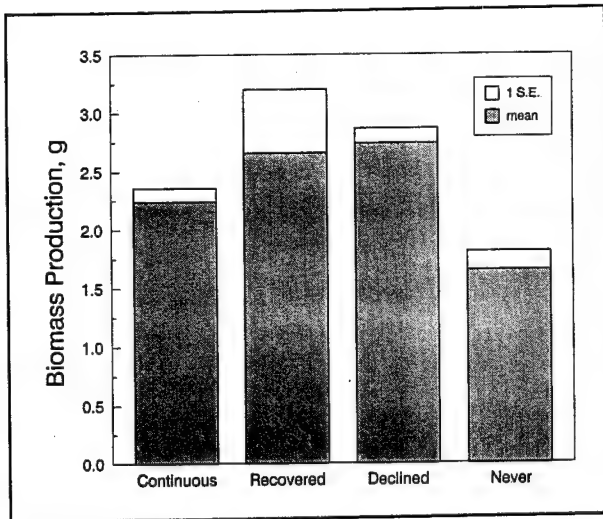


Figure 5. Growth of *M. Spicatum* plants on sediments from sites with differing *M. spicatum* cover histories. Cover history categories refer to sites where *M. spicatum* coverage from 1988 to 1993 was continuous, had declined and recovered (Recovered), declined and not recovered (Declined), or where *M. spicatum* had never occurred (Never)

availability (expressed as Secchi disk depth divided by water depth) varied significantly between areas of differing *M. spicatum* histories. Light availability was highest in continuously vegetated sites and was significantly lower in sites where vegetation had declined or was never present (Figure 6). Sites where vegetation had declined and recovered had an average light availability that was intermediate between that of continuously vegetated sites and decline sites.

Discussion

The results of plant growth bioassays indicate that sediment-related mechanisms, such as nutrient depletion or organic accumulation, probably did not contribute to the 1988-91 decline in submersed vegetation in TVA reservoirs. The absence of systematic sediment difference between decline and nondecline sites, coupled with the observed differences

in light regime between these areas, corroborate assertions that the decline probably resulted from effects of climatic factors that reduced light availability in 1989 through 1991 (Scott 1993). By using Secchi depth divided by water depth as measure of light regime, we were able to integrate the effects of differences in water clarity and depth. The integrated measure worked well to separate sites because many sites where *M. spicatum* populations had recovered typically had higher water clarity but were deeper than continuously vegetated sites.

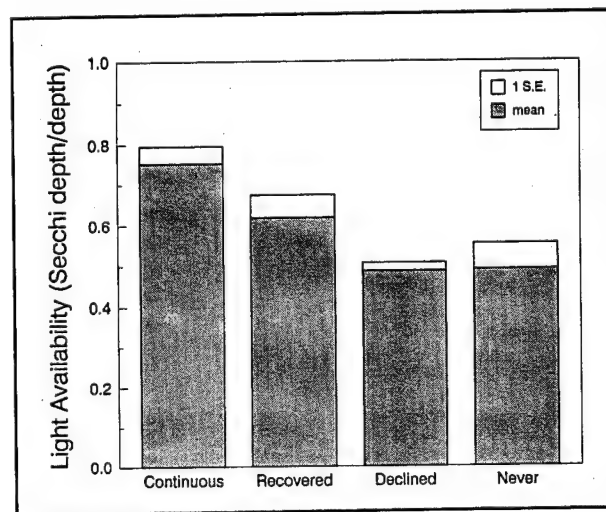


Figure 6. Light index for sites with differing *M. spicatum* cover histories (see Figure 5 for key to cover histories)

Other causes are possible, but most are not likely. The role of insects and pathogens was evaluated in separate studies coordinated with this one. None of the insect herbivores reported to impact *M. spicatum* was found in plant samples collected simultaneously with the samples used in this study.¹ The potential role of a pathogen or pathogen(s) in the decline is unclear. *Mycocleptodiscus terrestris*, a fungus which has been shown to impact *M. spicatum* in laboratory and greenhouse tests, was found in moderate numbers in plant samples from all reservoirs sampled.²

¹ Personal communication, M. Grodowitz, WES.

² Personal communication, J. Shearer.

One reason there has been so little success in attributing specific causal mechanisms to particular macrophyte declines is that many different phenomena, presumably having different causes, are lumped together as declines. Submersed macrophyte populations typically exhibit substantial fluctuations from year to year, and it is very difficult to separate short-term fluctuations from long-lasting reductions in coverage and biomass. I suggest categorizing declines based on their duration, scope, and geographical extent (Table 2), in order that different mechanisms can be identified for various classes of declines. The decline reported on here was relatively short-lived (i.e., about 3 years in duration) and coincided with declines in submersed vegetation over broad areas of the eastern United States. These characteristics suggest a climatic cause.

Table 2
Characteristics Used to Classify
Macrophyte Declines

Duration	Scope	Extent
Intra-annual	One species	One lake
Inter-annual	Several species	Several lakes
Several years	Entire type	Regional
Indefinite	Several types	

Declines of exotic submersed macrophytes are more likely to involve sediment-related mechanisms in natural lakes than in reservoirs. Reservoirs represent a dynamic sedimentation environment, with substantial potential for erosion and deposition. None of the samples evaluated in this study were the very fine, flocculent sediments that often accumulate under *M. spicatum* populations in lakes. In reservoirs, such sediments would likely be removed by erosion or altered by inorganic sedimentation. In lakes, such sediments often accumulate, leading to a poor substrate for plant growth (Barko and Smart 1986).

Declines caused by alterations of the sediment beneath exotic macrophyte populations would be expected to be local and relatively long-lasting. The decline of *M. spicatum* in

Lake Wingra from 1977 to 1978 (Carpenter 1980) had characteristics consistent with sediment-related causes, i.e., it was confined to several lakes which had supported *M. spicatum* populations for similar lengths of time and *M. spicatum* has not ever regained its former dominance in them. Future investigations of the contribution of sediment-related mechanisms to macrophyte declines should target such local, long-lasting declines.

Acknowledgments

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Sources of Mineral Nutrition for Submersed Macrophyte Growth in Riverine Systems: Results of Initial Investigations

by

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Biologists working on the Upper Mississippi River (UMR) have long been interested in submersed aquatic macrophyte populations because of their value to the river ecosystem. Submersed aquatic macrophyte communities provide food and shelter to a variety of organisms, influence nutrient dynamics, and stabilize sediments (Muencher 1944; Barko, Gunnison, and Carpenter 1991; Barko, Smart, and McFarland 1991; Poe et al. 1986)

In the UMR, recent declines in *Vallisneria americana* and other submersed species coincided with a prolonged drought. In just one backwater (Lake Onalaska), *Vallisneria* declined from greater than 1,200 ha to less than 120 ha during the late 1980s and early 1990s.⁴ Similar declines were reported from elsewhere in the UMR.⁵ Although factors affecting the declines were never positively identified, transplants of *Vallisneria* grew successfully in Lake Onalaska in 1992 (Rogers and Barko 1993). Since then, there has been an interest in restoring *Vallisneria* by means of large transplanting efforts. Nearly all parts of this macrophyte, especially tubers and rootstocks, are valuable as food to aquatic animals and migrating waterfowl (Korschgen, George, and Green 1988).

One likely factor limiting growth of submersed aquatic plant populations is nutrient availability. In order to sustain macrophyte growth from year to year in aquatic systems,

nutrient supplies to the sediments need to be replenished as they become depleted due to plant uptake and diffusional losses. Nutrients in various forms are transported via sedimentation (Forsberg 1989; James and Barko 1990). In concert with bioturbation, sediment mixing provides a primary source of nutrients to rooted submersed macrophytes (Barko, Gunnison, and Carpenter 1991). During prolonged drought (as occurred between 1987 and 1989), nutrient transport may be affected by low flows and an associated reduction in sediments to backwaters. In this study, we begin efforts to determine sources and mechanisms of nutrient replenishment, primarily nitrogen (N), in Lake Onalaska. These efforts are designed to provide a better understanding of the mechanisms of nutrient transport and renewal in the UMR. Here we present results of preliminary analyses of N concentrations in plant tissues, sediments, and water, and N deposition rates from accreted seston.

Study Site and Methods

The study was conducted during the summer of 1994 in Lake Onalaska (Pool 7) of the UMR System (Figure 1). This 2,835-ha lake is shallow (mean depth = 1.3 m) and has supported aquatic vegetation since it was formed by impoundment in 1937 (Green 1960). A bed of *Myriophyllum spicatum* and an adjacent open water site, approximately 680 yd from

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³ U.S. Army Engineer Waterways Experiment Station, Environmental Laboratory, Vicksburg, MS.

⁴ C. Korschgen et al., National Biological Service, La Crosse, WI, unpubl. manuscript.

⁵ J. Lennartson, U.S. Fish and Wildlife Service, Winona, MN; J. Wetzel, Wisconsin DNR, La Crosse, WI; and C. Korschgen, National Biological Service, La Crosse, WI; memorandum dated October 3, 1989.

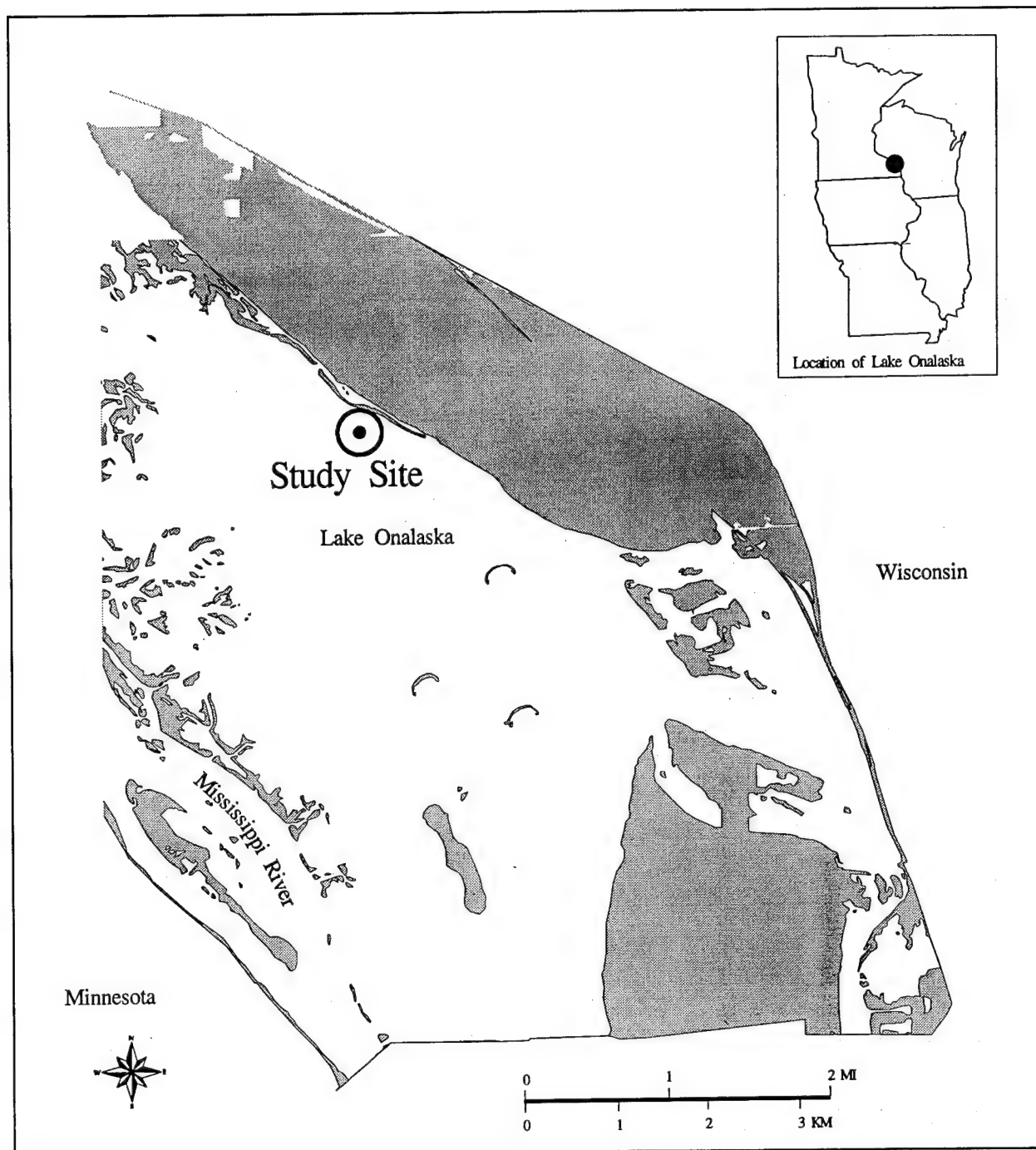


Figure 1. Location of study site in Lake Onalaska, Pool 7, of the UMR, during 1994

the north shore, were used as sites for the study. Aboveground biomass and tissue nutrients of *Myriophyllum* plants were monitored monthly. Five randomly placed quadrats (0.25 by 0.25 m) were used for harvesting plant tissues within the vegetated site. Plants were rinsed of loose epiphytic materials in the field and subsequently oven-dried at 80 °C

for 48 hr. Dried tissue samples were ground in a Wiley Mill and digested with sulfuric acid and hydrogen peroxide (Allen et al. 1974). Total N concentrations were determined colorimetrically with a Technicon Autoanalyzer II (Bran and Luebbe Analyzing Technology, Inc., Elmsford, NY), following methods of APHA (1985).

Sediment cores were taken approximately every 4 weeks (three cores at random points at each site). Cores were collected to a depth of 20 cm with a Wildco sediment sampler (Wildlife Supply Co., Saginaw, MI) equipped with an acrylic core liner (6.5 cm ID). The cores were sectioned in the field at 5-cm intervals. Each section was subsequently analyzed for exchangeable N, extractable P, moisture and density, and percent organic matter. Sediment moisture content and density were evaluated gravimetrically after oven-drying sediment at 105 °C for ~12 hr. Dried samples were combusted at 550 °C for estimations of organic matter content from loss of mass following ignition (Allen et al. 1974). Exchangeable $\text{NH}_4\text{-N}$ was obtained using a cation exchange procedure modified from Bremner (1965). Concentrations were determined colorimetrically with a Technicon Autoanalyzer II.

Seston accretion was qualitatively assessed at vegetated and open sites using sediment traps. Each trap consisted of two or three (3-in. ID by 12-in.) PVC cylinders and a holding carousel. Contents of traps were collected at approximately 10-day intervals and were analyzed for total dry mass, ash weight, and N following procedures described in James and Barko (1993) and James, Kennedy, and Gaugush (1990).

To examine spatial and temporal changes of N in sediment pore water and overlying bottom waters, Plexiglas peepers, similar to those described in James and Barko (1991), were placed within the vegetated and open sites once a month. The 12 chambers in each peeper were filled with deoxygenated water and were covered with nucleopore dialysis membranes (0.4- μm pore size). Peepers were placed in sediments at both sites and were left in place to equilibrate for 10 to 14 days. Several chambers of each peeper were left above the sediment-water interface to sample water column nutrients. Water samples from the peeper chambers were withdrawn with syringes immediately following peeper retrieval and were placed in prepared vials. Samples were analyzed for $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{+NO}_2\text{-N}$.

Results

Aboveground biomass averaged 92.89 g/m^2 at the beginning of the study and reached a maximum in mid-July, estimated at 302 g/m^2 . Total nitrogen (TN) concentrations of aboveground plant tissues were well above critical levels required for healthy growth (Gerloff and Kromholz 1966), averaging 3.31 percent in aboveground plant tissues (Table 1).

Table 1
Mean Aboveground Tissue Biomass Data and Associated TN Concentrations in *Myriophyllum spicatum* Bed, Lake Onalaska, Pool 7, UMR, 1994

Month	Biomass, g/m^2	Total Nitrogen, mg/g
June	92.8 (15.7)	32.88 (0.55)
July	302.1 (64.2)	31.92 (0.63)
August	143.6 (25.9)	33.34 (0.86)
September	36.7 (9.6)	34.12 (0.41)

$\text{NH}_4\text{-N}$ concentrations collected from the peeper chambers were relatively stable over time but varied with depth (Figure 2, top). Seasonal means were consistently higher in chambers below 5 cm both in the vegetated site and the open site, averaging (between 8 and 13 cm below the surface) 4.47 mg/L ($n = 8$, standard error = 1.0 mg/L) at the open site and 2.63 mg/L ($n = 9$, standard error = 0.08 mg/L) at the vegetated site.

$\text{NO}_3\text{+NO}_2\text{-N}$ concentrations at each depth were generally more variable than $\text{NH}_4\text{-N}$ over time. Seasonal means in chambers above the sediment surface were significantly higher (t test, $P < 0.05$) than in pore water at both sites (Figure 2, bottom). Concentrations in chambers above the sediment surface revealed $\text{NO}_3\text{+NO}_2\text{-N}$ averaged 0.60 mg/L at the open site ($n = 47$, standard error = 0.03 mg/L) and 0.62 mg/L at the vegetated site ($n = 55$, standard error = 0.04 mg/L).

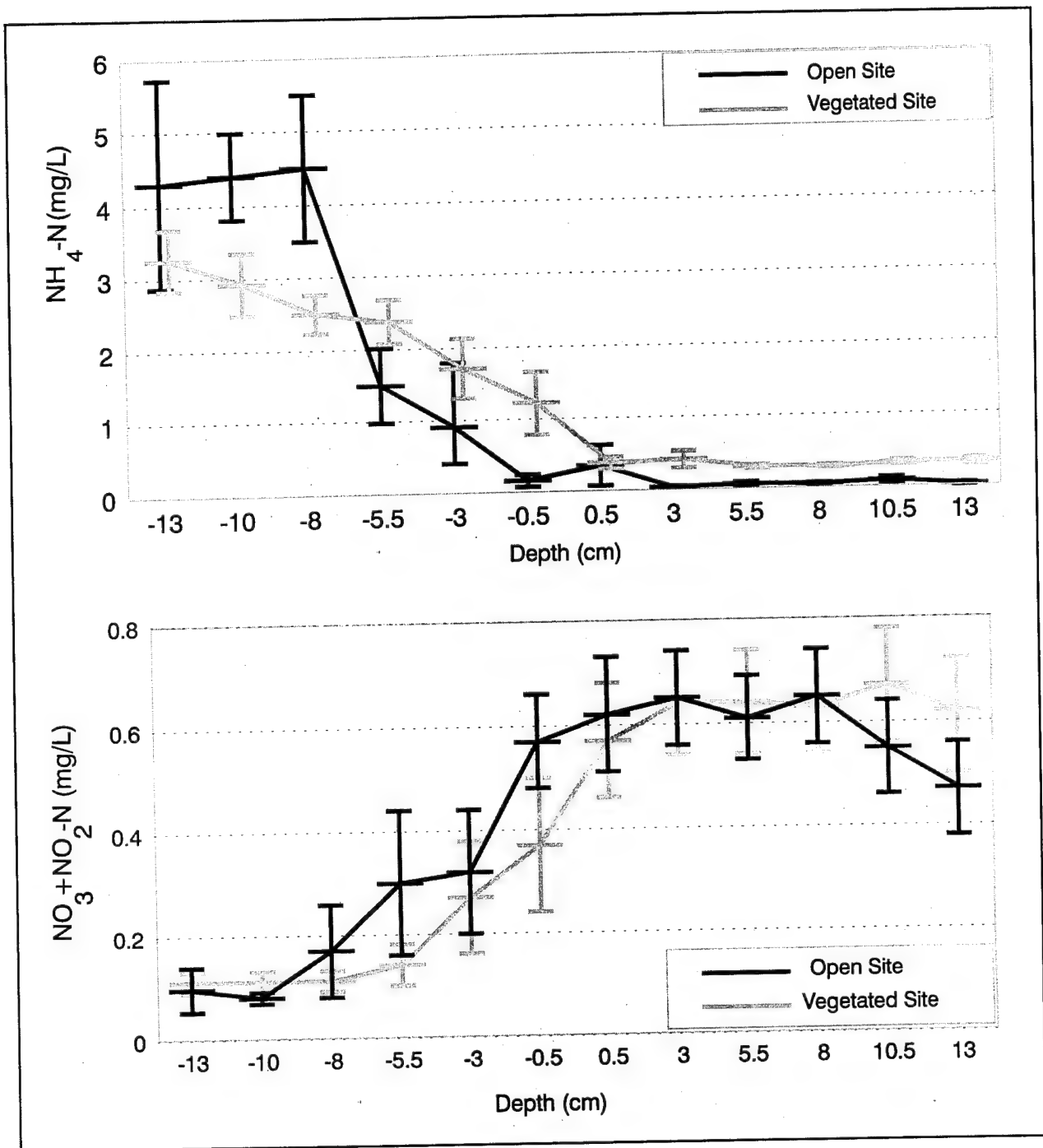


Figure 2. Seasonal means of $\text{NH}_4\text{-N}$ and $\text{NO}_3 + \text{NO}_2\text{-N}$ concentrations in pore water, collected from peeper chambers in Lake Onalaska, 1994. Depths relate to location of the chambers in centimeters above or below the sediment-water interface. Means and standard errors are based on six to nine determinations at each depth

Mean sediment density, moisture, organic content, and $\text{NH}_4\text{-N}$ concentrations were not significantly different (t test, $P > 0.05$) between the vegetated and nonvegetated sites and thus were averaged across sites (Table 2).

$\text{NH}_4\text{-N}$ and organic matter were all significantly higher ($P < 0.05$) in the sediments below 10 cm than in the upper 10 cm. However, moisture and density did not vary vertically throughout the profile.

Table 2
Mean Physical and Chemical Characteristics ± 1 SE (N = 24) of Sediments

Depth, cm	Moisture, %	Density, mg/L	Organic, %	NH ₄ -N, mg/g
1-5	4.87 \pm 0.58	0.39 \pm 0.6	1.52 \pm 0.25	0.01 \pm 0.00
6-10	4.20 \pm 0.50	0.49 \pm 0.04	1.83 \pm 0.16	0.02 \pm 0.00
11-15	4.89 \pm 0.52	0.45 \pm 0.06	3.05 \pm 0.35	0.03 \pm 0.01
16-20	4.76 \pm 0.51	0.46 \pm 0.05	3.68 \pm 0.48	0.05 \pm 0.01
21-25	4.78 \pm 0.60	0.48 \pm 0.05	3.83 \pm 0.83	0.05 \pm 0.01

Note: Data from vegetated site and unvegetated site are combined.

Sediment trap data indicate Total N deposition varied throughout the season. Deposition rates were highest in June and early July, and lowest in early August. Between sites, significant differences (*t* test; $P < 0.05$) in rates occurred during most time periods after mid-July (Figure 3). Differences in deposition rates between sites during these time periods may have been due to the effect of plant bed structure on hydrological conditions. Total N deposition rates for the season averaged 580.72 mg/m²/d at the open site ($n = 23$, standard error = 42.77 mg/m²/d) and 448.92 mg/m²/d at the vegetated site ($n = 23$, standard error = 30.76 mg/m²/d). These mean deposi-

tion rates for N were significantly different (*t* test, $P < 0.05$) between the two sites.

Discussion

To date, information has not been available to identify sources and mechanisms of N replenishment to backwaters of the UMR. While we are not yet at a point where we can construct a budget for Lake Onalaska, our early analysis of N data presented in this paper indicates potential sources of this nutrient. It is clear that *Myriophyllum* plants monitored throughout this study were able to maintain

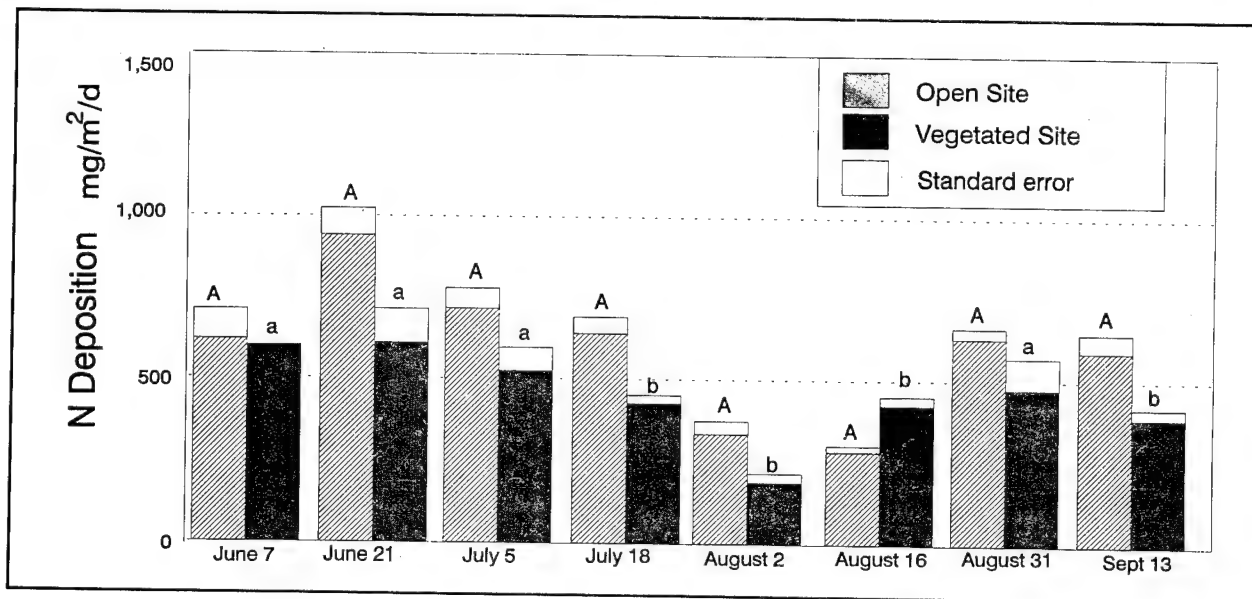


Figure 3. Total N deposition collected in sediment traps at the open water and vegetated site, Lake Onalaska, 1994. Means and standard error bars are based on three replicate determinations. Within each date, deposition rates sharing the same letter do not differ significantly from each other

adequate amounts of N for growth. Levels of N supply to the plants were therefore considered to be nonlimiting. Concentrations of N in the form of $\text{NH}_4\text{-N}$ were higher in the sediment pore water than was any other form of N in the sediments or in the water column. Thus, it appears that sediment pore water $\text{NH}_4\text{-N}$ may provide a major source of this nutrient to the plants. The importance of sediments as a direct source of N for aquatic macrophytes is ecologically significant because rooted aquatic macrophytes rely primarily on nutrient uptake by roots (Barko, Gunnison, and Carpenter 1991) and, in general, can obtain nutrients exclusively from sediments, even in riverine systems with fast flowing currents (Chambers et al. 1989).

Relatively low concentrations of N in the water column compared with N concentrations in sediment have been recognized in other systems (Barko, Gunnison, and Carpenter 1991). Thus, similar findings were not unexpected for this backwater. Even more interesting, however, is our sediment trap data which indicate that N deposition via accretion may provide an important means of replenishment to the sediment surface. Barko and Smart (1986) suggested that growth of submersed macrophytes may be stimulated by increased sediment fertility due to sedimentation of fine-textured inorganic materials. Results of leaching studies we conducted in the laboratory with the sediment trap contents (currently under analysis) will provide a better indication of potential N release rates to each site under both anaerobic and aerobic conditions. At least in Lake Onalaska, this method of N renewal, combined with processes of sediment mixing, may prove to be an important mechanism in maintaining aquatic macrophyte populations in sediments with high sand fractions (Rogers, McFarland, and Barko 1995).

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Revegetation and Propagule Transport in the Tidal Potomac River

by

Nancy Rybicki¹ and Virginia Carter¹

Introduction

A resurgence of submersed aquatic macrophytes in the tidal Potomac River in 1983 (Carter and Rybicki 1986) was followed by several years of population expansion in shallow-water areas (<2 m) (Carter et al. 1994). In 1989, macrophyte populations declined in the upper tidal river above Marshall Hall but continued to expand in the lower tidal river (Marshall Hall to Quantico) (Figure 1). Macrophyte populations in the upper tidal river had failed to recover by 1993 and, more recently, there has been a decline in macrophyte populations in the lower tidal river as well.

By 1988, the dominant plant species in both the upper and lower tidal river was a monoecious strain of the southeast Asian exotic *Hydrilla verticillata*, which grows in dense beds in shallow areas and generally outcompetes other macrophyte species. The decline in macrophytes was largely attributed to the disappearance of the dense beds of *H. verticillata* (Carter et al. 1994). At present (1994), a few dense beds of *H. verticillata* remain in the upper tidal river in addition to a larger population in the lower tidal river. Populations of four other species, *Myriophyllum spicatum*, *Vallisneria americana*, *Ceratophyllum demersum*, and *Heteranthera dubia*, continue to be found in moderate abundance in the tidal river, and other species, such as *Potamogeton pectinatus*, *Potamogeton crispus*, *Zannichellia palustris*, *Najas marina*, and *Najas guadalupensis*, are present in small, isolated beds or mixed with the more dominant species.

We have suggested that light is the most important factor controlling the distribution of submersed macrophytes in the tidal Potomac River (Carter et al. 1994). However, other factors may be involved in the failure of some sites to become vegetated during the resurgence and subsequent spread of macrophytes in shallow water after 1983, and in the failure of previously vegetated sites to become revegetated after the disappearance of vegetation in 1989. One possible factor is the lack of propagules of various species; if plant propagules failed to reach the unvegetated sites, adequate water clarity would not result in revegetation.

In the tidal Potomac River, submersed macrophytes overwinter in a dormant state and reproduce both vegetatively and by seed during the growing season. Some species (*H. verticillata*, *V. americana*, and *P. pectinatus*) overwinter by means of tubers buried in the sediment while others (*H. verticillata*, *P. crispus*) overwinter by winter buds or turions which fall to the surface of the sediment. A few species (*M. spicatum* and *H. dubia*) overwinter as rootstocks and stems which sprout in the spring.

For many species, vegetative reproduction is the most common and successful mode of population expansion (Sculthorpe 1967; Hutchinson 1975). Vegetative reproduction includes expansion of parent plants by runners or rhizomes which root at the nodes and form new plants. Revegetation is also possible from plant fragments that grow roots rapidly from nodes and are carried up- and downriver by tidal currents and wind. All the species in the tidal Potomac, except for *H. verticillata*, produce seeds annually; however, little is

¹ U.S. Geological Survey, Reston, VA.

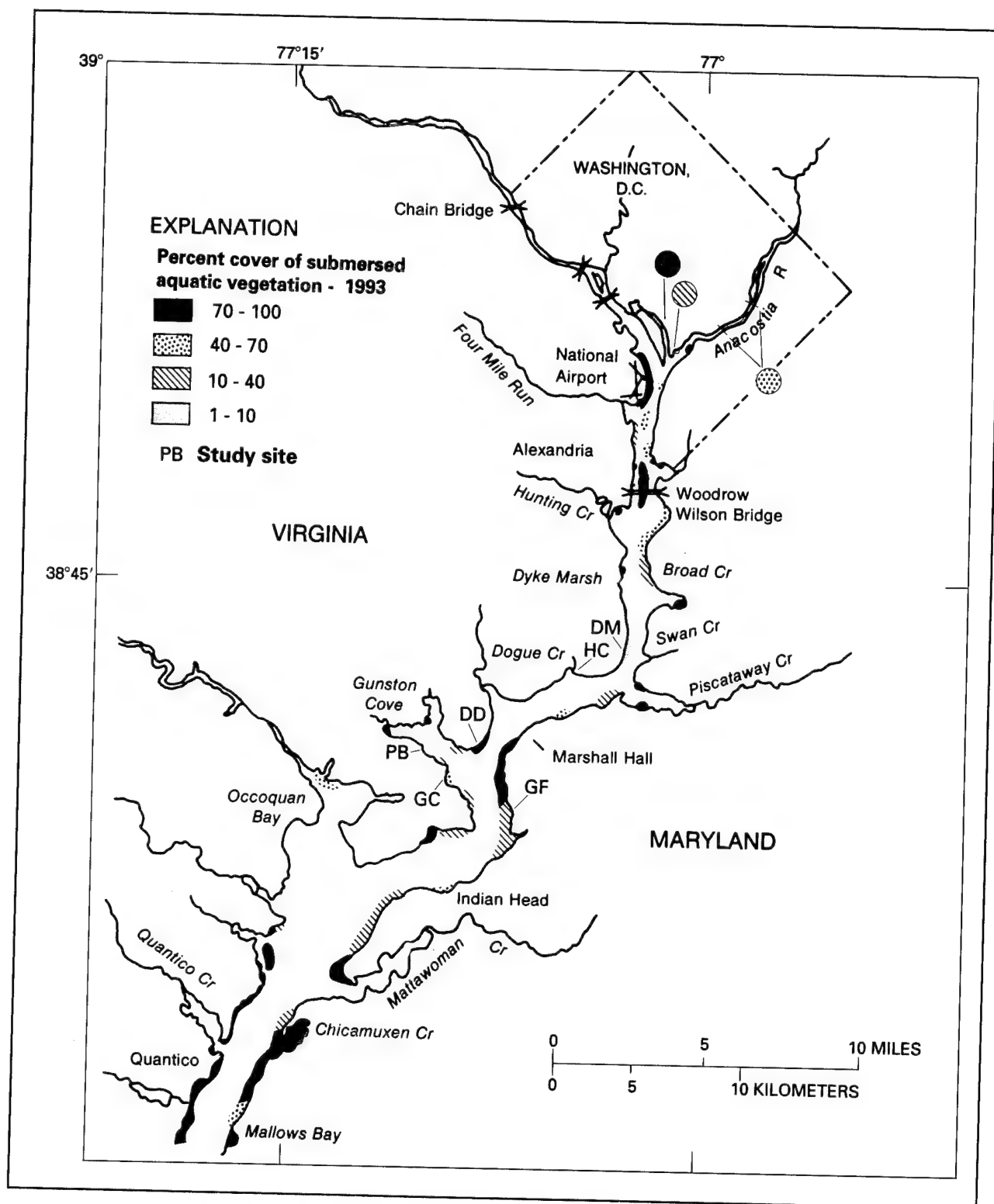


Figure 1. Map of the tidal Potomac River showing 1994 sampling sites and 1993 areal coverage of submersed aquatic vegetation in the tidal Potomac River

known about the viability of the seeds from many of these plants. Seeds are buoyant, but their weight and shape tend to cause individual seeds to fall close to the mother plant (Orth,

Luckenbach, and Moore 1994). Seeds may also be transported in small numbers by waterfowl (Vivian-Smith and Stiles 1994). Seed heads, tubers, and turions attached to plants

uprooted by wind and current may float for relatively long distances before settling to the sediment.

The purpose of this study was to measure the pregrowing- and postgrowing-season availability of propagules and the flux of macrophyte propagules at unvegetated sites during the growing season (April to October). Our primary hypothesis was that there was adequate plant material moving through the unvegetated sites to result in revegetation if water clarity and other factors were favorable. We further hypothesized that pre-season distribution of propagules combined with water clarity measurements could be used to predict occurrence of species and biomass trends at a site. We therefore measured pre- or post-season availability of propagules at three nearby vegetated sites for comparison with unvegetated sites, sampled growing-season species composition and biomass at all sites, and made regular light attenuation and water quality measurements.

Study Sites

Three unvegetated sites (PB, HC, and DM) and three vegetated sites (GF, DD, and GC)

were chosen in the freshwater tidal Potomac on the basis of areal coverage in the 1993 growing season (Figure 1) and the period from 1988 to present. Site GC was added as an additional vegetated site during the summer. All sites were in the freshwater tidal Potomac River. Table 1 shows the status (vegetated or unvegetated) of each site and the areal coverage and species composition in 1988 and 1993.

Methods

Preseason and postseason sampling

During the periods before plant germination (March) and after the growing season (November, December), the sites were sampled for tubers, turions, and plant fragments remaining from the previous year's growing season. Five sites were sampled in March and six sites (GC was added) were sampled in November. At each site, 20 cores were taken with a posthole digger (182 cm²) at water depths of 0.5, 1.0, 1.5 m at approximately low tide. Posthole digger cores were about 10 cm deep but ranged from 10 to 20 cm depending on substrate composition.

Table 1
Status of Each Site (Vegetated or Unvegetated), Areal Coverage, and Species Composition in 1988 and 1993

Site	Status	Areal Coverage (1988 to 1993)	Percent Areal Coverage in 1988 or 1993	Species Composition in 1988	Species Composition in 1993
PB	Unvegetated	None	0 ('88); 0 ('93)	None	None
HC	Unvegetated	Decreased	100 ('88); 0 ('93)	<i>H. verticillata</i>	None
DM	Unvegetated	Decreased	100 ('88); < 10 ('93)	<i>H. verticillata</i>	Trace <i>H. verticillata</i> and <i>Potamogeton pectinatus</i>
GF	Vegetated	Decreased	100 ('88) 40 to 70 ('93)	<i>H. verticillata</i>	<i>Myriophyllum spicatum</i> (80%); <i>H. verticillata</i> (20%)
DD	Vegetated	Unchanged	40 to 70 ('88) 40 to 70 ('93)	<i>H. verticillata</i> (60%); <i>V. americana</i> (30%); <i>M. spicatum</i> (5%); <i>Heteranthera dubia</i> (5%)	<i>V. americana</i> (80%); <i>M. spicatum</i> (20%)
GC	Vegetated	Unchanged	40 to 70 ('88) 40 to 70 ('93)	<i>M. spicatum</i> (80%); <i>H. verticillata</i> (14%); <i>V. americana</i> (5%); <i>H. dubia</i> (1%)	<i>H. verticillata</i> (50%); <i>V. americana</i> (40%); <i>M. spicatum</i> (10%)

Twenty grab samples for plant fragments, turions, and tubers near the sediment surface were collected at 0.5-, 1.0-, 1.5-, 2.0-m depths using modified oyster tongs (surface area = 900 cm²). At the 2-m depth, the oyster tongs were pushed well into the sediment for 10 of the grabs (changed to five grabs for the postseason sampling) to collect samples equivalent to those made with the posthole diggers. The oyster tongs do not collect as deep a sample as the posthole diggers, and tubers >10 cm deep in the sediment, if any, would have been missed. The samples were sieved through 1.5-mm-mesh screen. We were not always able to distinguish between *H. verticillata* tubers and turions, so these were combined and counted together, and were referred to throughout as tubers.

Hydrilla verticillata tubers and turions were stored in the refrigerator at 6 °C and sent in a cooler within 5 days to the Environmental Laboratory, WES, where they were chilled for approximately 3 weeks, separated into weight classes, planted, and observed for germination over a period of 3 weeks (McFarland and Barko 1995).

Tubers of *V. americana* and *P. pectinatus* were planted and observed for germination and viability over a period of 3 weeks in the U.S. Geological Survey (USGS) lab in Reston, VA. In Reston, viability of plant fragments other than *H. verticillata* was tested by planting tubers or fragments in small plastic containers (12 by 7 by 5 cm deep) filled with sediment from the PB site and submersed in tubs filled with a low-alkalinity culture solution containing major nutrients except for nitrogen and phosphorus to minimize algae growth (Smart and Barko 1985). Water temperature was approximately 25 °C. Tubers were aerated and artificial light was provided for 12 hr/day. In the postseason sampling, tubers were chilled for approximately 3 weeks to simulate a dormant winter period. *Vallisneria americana* tubers were separated into size classes based on length. Tubers were considered viable if they sprouted and grew recognizable leaves. Plants were considered viable if they increased in length and were green and healthy in 3 weeks time.

Preseason sampling for seeds at vegetated and unvegetated sites (DM, HC, DD, PB, and GF) was conducted during the period March 8-25. Ten 10-cm-deep sediment samples were collected with posthole diggers in water 0.2 to 1.0 m deep. Sediment was sealed in plastic bags and kept chilled for a maximum of 4 days. A subsample of sediment from each bag was transferred to containers similar to those used for viability and placed in a tub of culture solution at 25 °C. Tubers were aerated, and artificial light of 290 $\mu\text{E m}^{-2} \text{sec}^{-1}$ was provided for 14 hr/day for 3 to 6 weeks; any seedlings were recorded after 3 weeks.

Ten liters of sediment was collected from each of the three unvegetated sites (PB, DM, and HC) in March in water 0.2 to 1.0 m deep. The sediment was kept chilled and sent to WES in coolers within 5 days. The sediment texture and nutrient composition was determined, and the sediment was used for *H. verticillata* growth experiments (bioassays) (McFarland and Barko 1995). Growth responses were compared for *H. verticillata* grown on sediment from Brown's Lake, WES (reference sediment), and from the three unvegetated sites (McFarland and Barko 1995).

Growing season

During the growing season, transport and deposition of propagules were measured at sites DM, PB, and HC from May to September using three data collection methods. The first method was used only in the intertidal zone in May, when the plants were beginning to germinate. In the intertidal zone, the number and species of plants in 2.3-m² quadrats were recorded at each site. The second method was used to measure propagule transport through the site using nets. Three nets (1 by 0.6 m) made from 10- by 10-cm wire fencing covered with 1- by 1-cm plastic netting with bouys attached for flotation were set out randomly at each site at low-tide depths between 0.2 and 1.0 m. Nets were hung on poles and allowed to float up and down with the tide, so that they collected fragments floating at the surface to 0.6 m below the surface.

The third method was used to measure propagule deposition throughout the growing season. Traps were constructed of 0.6- by 0.6-m pieces of hardware cloth. Twelve of these were placed on the sediment at each site. Two types of hardware-cloth traps were used. One type was flat with a piece of fine screen covering the top side. Traps made of this material were intended to catch plant fragments and tubers that might fall off the fragments. The other type had ten 10-cm wide by 10-cm tall hardware cloth "prongs" sticking up vertically to entrap the fragments rolling along or settling on the bottom. Ropes were attached on all four sides, and traps were anchored to the sediment with 1-cm-diam poles pushed through two holes, one on each side of the trap. The traps were pulled up by hooking the attached ropes and pulling the trap gently upward along the poles. During May, nine flat traps and three traps with prongs were set out to test the two designs. It was determined that the tubers moving through the site were well attached to the plant fragments, and on June 7, all traps were converted to prong traps.

Traps and nets were left in the river for intervals of 4 to 11 days. When the nets and traps were pulled up, all fragments of vegetation were collected and sorted by species. *Hydrilla verticillata* fragments and any attached tubers and or turions were kept chilled and mailed to WES. WES personnel counted the number of fragments and measured the wet weight of the *H. verticillata* on each trap. A conversion factor was determined to convert wet to dry weight (0.07 by wet weight). Fragments of other species were returned to the USGS laboratory. From May 9 through June 22, the fragments of species other than *H. verticillata* were counted but not weighed. Fragment viability was tested at the USGS and WES by planting fragments in sediment using the same methods used for tuber viability studies. Beginning on June 30, fragments of species other than *H. verticillata* were dried in an oven for 10 to 12 hr at 105 °C, and biomass was measured in grams dry weight (gdw).

Flux was calculated for *H. verticillata* and other species by converting individual dry

weights for each species on each trap to gdw m^{-2} and then dividing by the number of days the traps were deployed. Mean fluxes were compared using an analysis of variance test, and statements of statistical significance refer to probability levels of 5 percent or less.

Transects were set up perpendicular to shore from the high tide mark at both vegetated and unvegetated sites, and grab samples were collected with oyster tongs at stations established at 10-m intervals. Three grabs were made at each 10-m station, and samples were collected to 60 m from shore. If vegetation was present at 50 or 60 m, sampling was continued at 10-m intervals until all grab samples at two consecutive stations contained no vegetation. Vegetation was sorted by species, dried overnight at 105 °C, and weighed to give gdw; sample biomass was converted to gdw m^{-2} . To normalize biomass data to compare sites, only biomass collected in the first 60 m of the transects was used. An average in gdw m^{-2} for each or all species was calculated by dividing the total biomass in 60 m by 18 (the total number of grab samples in the first 60 m from shore).

During July and August, we conducted an experiment to see how long fragments of *H. verticillata* could float in the river and remain viable. On July 6, 15-cm-long apical stems of *H. verticillata* were collected from the bed at GC. Fragments were placed in cylindrical cages (60 cm long and 15 cm in diameter) covered with 0.5- by 0.5-cm mesh with bouys for flotation. Cages were suspended in the river for treatment periods of 1, 2, and 4 weeks. At 0, 1, 2, and 4 weeks, plants were tested for viability (20 fragments per treatment) and analyzed for nitrogen, phosphorus, and carbohydrate content (three samples per treatment) at WES. Also, at 0, 1, 2, and 4 weeks, four sets of 10 fragments each were placed in flats on the bottom of the river at both 0.5- and 0.8-m depths at site DM to see if they would root. Flats consisted of 0.5- by 0.5-cm mesh bag held in shape by a 20- by 60-cm wire fencing. Two sets of plants at each depth were checked for roots at the end of 2 and 4 weeks.

Photosynthetically active radiation (PAR), Secchi depth, total suspended sediment (TSS), chlorophyll-*a*, and temperature were measured every 2 weeks at sites DM, HC, DD, and PB. The USGS and the Virginia Department of Environmental Quality (DEQ) sampled on alternate sampling dates. At two sites (PB and HC), DEQ sampled 500 m upstream of the sampling site, and at site HC, measurements were made 1 to 2 days later than the other sites and PAR was not measured. At sites DD and DM, DEQ and USGS sampled at exactly the same site. Water samples for TSS and chlorophyll-*a* were collected 0.3 m below the surface (analytical method according to U.S. Environmental Protection Agency 1982.) PAR was measured in air and below the surface simultaneously with a LICOR¹ 185B quantum sensor. Measurements were made just below the surface and at 0.5-m intervals until the reading fell below 10 $\mu\text{E m}^{-2} \text{sec}^{-1}$. Light attenuation coefficients (K) were calculated using Beer's Law

$$I_z = I_0 e^{-Kz}$$

where I_z is light measured at depth z , and where I_0 is light (PAR) measured by the surface sensor and corrected for reflection at the air-water interface by subtracting 6 percent of surface light.

Results

Preseason and postseason sampling

Preseason sampling showed that viable plant propagules existed at the two vegetated sites, GF and DD, and were predominantly *V. americana* tubers and *H. verticillata* tubers (Figure 2). At site DD, a few *H. verticillata* tubers were found at the 0.5-m depth and none at deeper stations, whereas at site GF *H. verticillata* tubers were found at all depths, with most 1.5 m deep. A few *V. americana* tubers were found at the 0.5-m depth at

site GF and none at deeper stations, whereas the number of *V. americana* tubers at site DD was greatest at 0.5 m and decreased with depth. Fragments (short stems) of *C. demersum* and *M. spicatum* were found at the 0.5- and 2.0-m depths at both sites. One plant fragment was found at sites HC and PB.

Viability studies conducted at WES showed that 67 percent of *H. verticillata* tubers germinated and grew (McFarland and Barko 1995). Percent germination increased with size class of tubers, all of which were from the GF site (McFarland and Barko 1995). Table 2 shows the preseason viability of plants other than *H. verticillata*. Viability for tubers and fragments was generally high (>90 percent) except for *V. americana* tubers at site GF (50 percent); however, the number of tubers tested at that site was very small.

The postseason distribution of propagules differed from the preseason distribution in terms of both numbers and distribution (Figure 2). In comparison with the preseason sampling, *H. verticillata* tubers were far fewer and were concentrated in shallower water at site GF, were sparse again at site DD, and were abundant at the new site, GC, but only at the 0.5-m depth. A few *H. verticillata* tubers were found at unvegetated site DM. Tubers of *V. americana* had decreased in number at site DD relative to preseason sampling, were not found at site GF, and were relatively abundant at site GC in shallower water. Two *P. pectinatus* tubers were found at site DM. At site GC, 95 percent of tubers were viable, and at site DM, 50 percent of the tubers were viable (McFarland and Barko 1995). Viability of propagules from plants other than *H. verticillata* is shown in Table 2. Both *P. pectinatus* tubers were viable. The *V. americana* tubers had an overall viability of 60 percent, but size classes showed a marked difference in viability: small tubers (10 to 20 mm) were only 35 percent viable (6 of 17) whereas large tubers (21 to 24 mm) were 100 percent viable (9 of 9).

¹ Use of the brand name in this report is for identification purposes only and does not constitute endorsement by the U.S. Geological Survey.

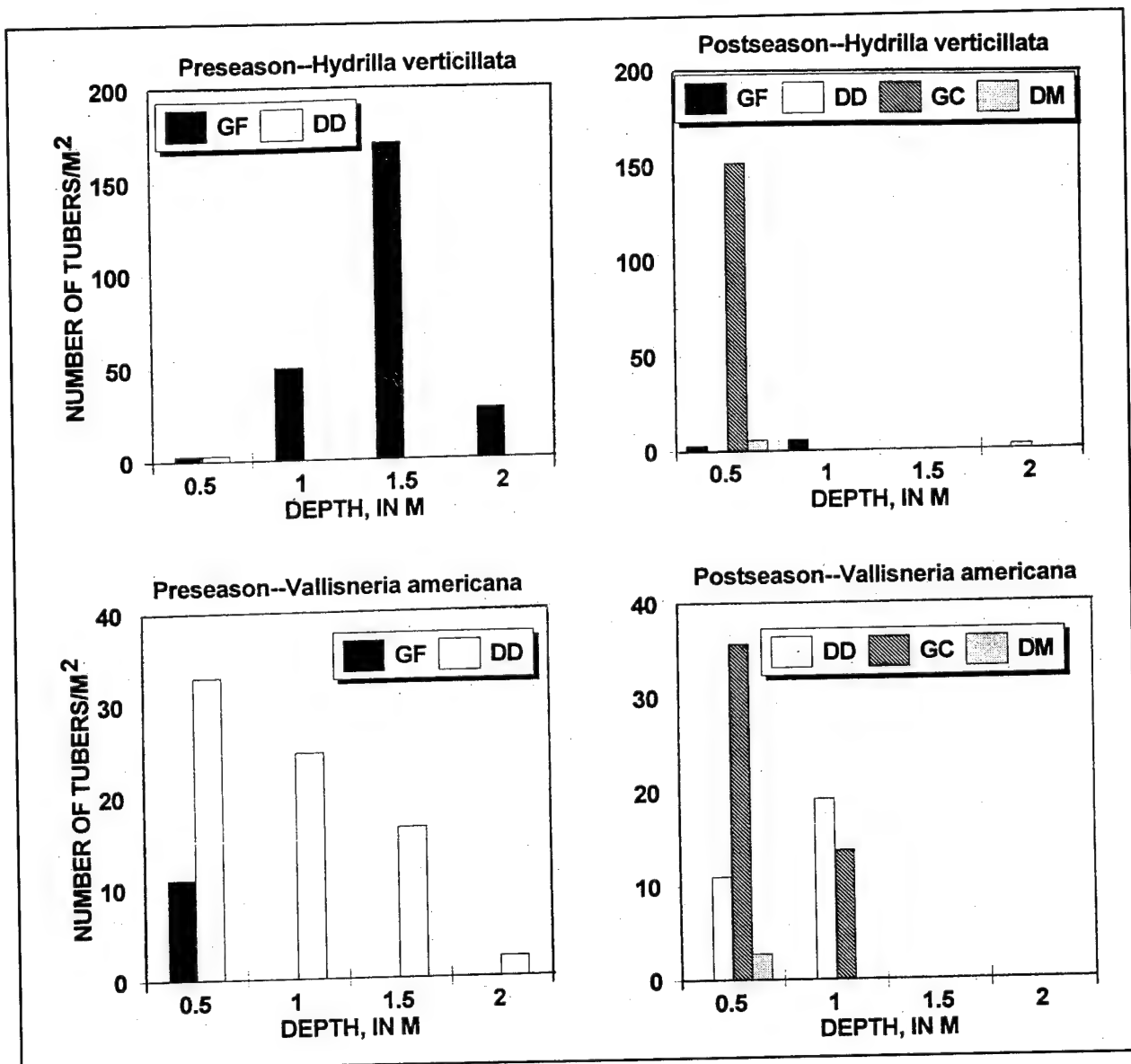


Figure 2. Preseason and postseason distribution of *V. americana* tubers and *H. verticillata* tubers and turions combined at vegetated and unvegetated sites in the tidal Potomac River

Preseason sampling for seeds suggested that a large number of seeds cannot be found at any of the sites in the spring. Only three seeds germinated in the laboratory, and one of these germinated in the fifth week, well after our 3-week limit. There was one *V. americana* sprout from sites HC and DM and one *Najas minor* sprout from site DD.

The bioassay on the sediment from the three unvegetated sites showed that *H. verticillata* achieved the most biomass on site PB

sediment and the least biomass on site HC sediment (McFarland and Barko 1995). After 5 weeks, plants grew from an initial height of approximately 10 cm to mean heights of 27, 32, and 37 cm at sites HC, DM, and PB, respectively. Plant biomass and height were significantly correlated with concentrations of exchangeable nitrogen, and growth responses on sediments from the unvegetated sites were, in nearly all cases, significantly less than those on the control sediment from Brown's Lake (McFarland and Barko 1995).

Table 2
Viability of Plant Propagules Other than *H. verticillata* Collected at Study Sites in the Tidal Potomac River During Preseason and Postseason Samplings

Species	Site	Plant Form	n	Percent Viable
Preseason				
<i>Vallisneria americana</i>	DD	Tuber	27	93
	GF	Tuber	4	50
<i>Myriophyllum spicatum</i>	GF	Stem fragment	10	100
	HC	Stem fragment	1	100
<i>Ceratophyllum demersum</i>	GF	Stem fragment	10	90
	PB	Stem fragment	1	100
Postseason				
<i>Potamogeton pectinatus</i>	DM	Tuber	2	100
<i>Vallisneria americana</i>	GC	Tuber	15	60
	DD	Tuber	10	60
	DM	Tuber	1	0

Growing season

Some propagules are deposited in the intertidal zone. Some newly germinated plants, mostly *H. verticillata*, were found in the intertidal zones of all three unvegetated sites in May (Table 3). We later observed that these plants did not survive through the end of June because of exposure at low tide and being dislodged by wave action in this zone. Plants in the intertidal zone probably sprouted from tubers, turions, or seeds deposited the previous fall when wind-driven mats of senescing plants were washed up on the shoreline.

Results from analysis of nets and traps deployed at the unvegetated sites, combined with frequent observations of shoreline detritus at low tide, showed that viable propagules of several species were washing into each site; the most propagules were deposited on the bottom in site DM and the least in site HC. Fragments from seven species were collected on the traps and nets combined at site DM during the period May 19-June 22 (Figure 3). The number of fragments of *H. verticillata* increased steadily during this period. Smaller numbers of fragments of other species were found; the highest species

Table 3
Number of Plants Found in the Intertidal Zone in May at Sites HC, PB, and DM in the Tidal Potomac River

Site/Date	Shoreline Area Sampled (m ²)	Number of Plants		
		<i>Hydrilla verticillata</i>	<i>Potamogeton pectinatus</i>	<i>Zannichellia palustris</i>
HC (5/2)	9.2	17	0	0
PB (5/2)	9.2	1	0	0
DM (5/2)	9.2	21	2	4
HC (5/26)	6.9	25	0	0
DM (5/26)	9.2	36	0	0

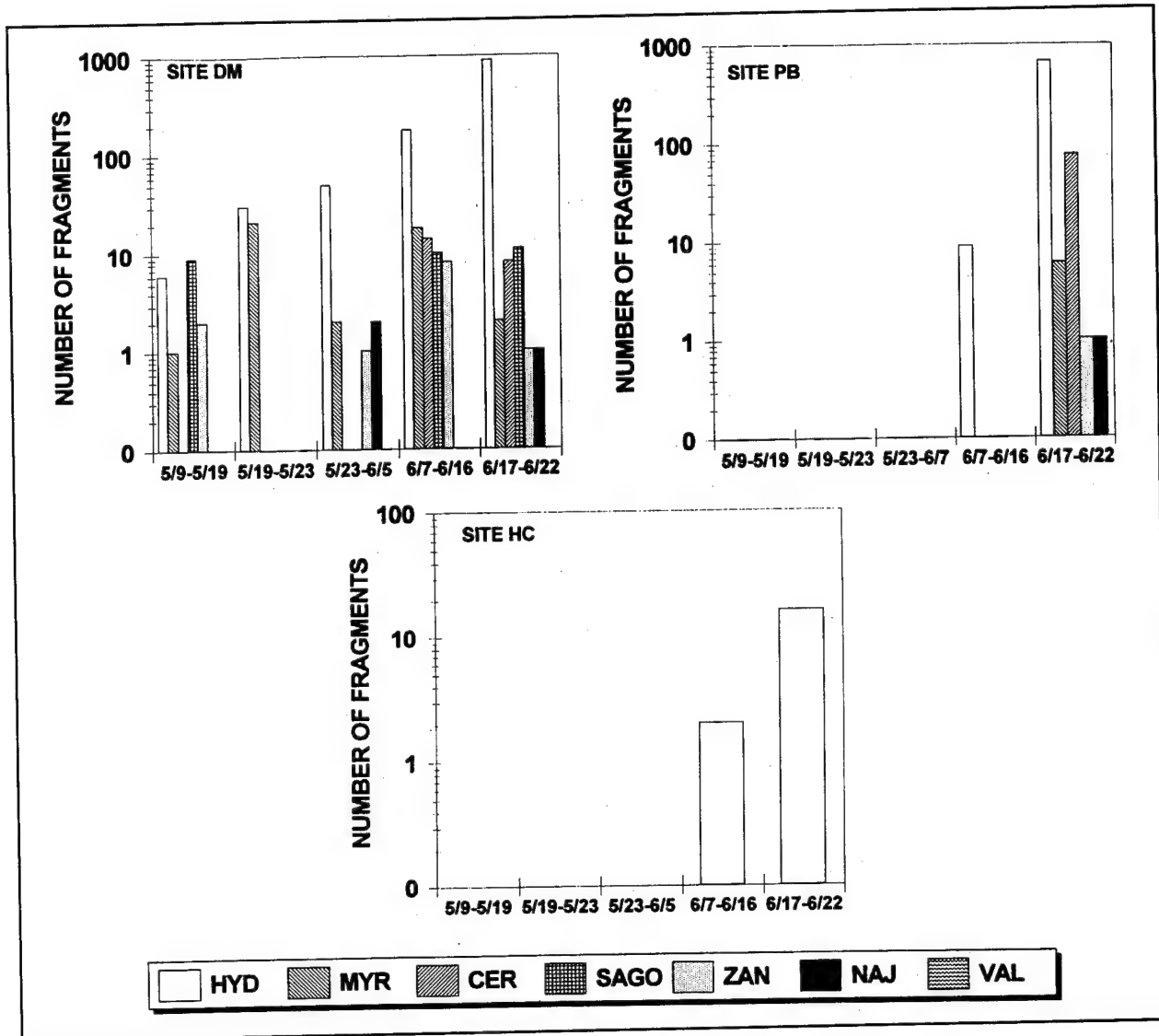


Figure 3. Number of fragments caught on traps and nets combined at unvegetated sites between May 19 and June 22, 1994. Dates indicate the number of days nets and traps were left in place. (HYD = *Hydrilla verticillata*, MYR = *Myriophyllum spicatum*, CER = *Ceratophyllum demersum*, SAGO = *Potamogeton pectinatus*, ZAN = *Zannichellia palustris*, NAJ = *Najas minor*, VAL = *Vallisneria americana*)

diversity occurred in June. No fragments were collected on the traps at site PB until June when fragments of five species were collected; *H. verticillata* and *C. demersum* were the primary species (Figure 3). The only species found at site HC was *H. verticillata*. The viability of fragments of various species found at site DM during the growing season was ≥ 70 percent (Table 4).

The mean biomass flux ($\text{gdw m}^{-2} \text{day}^{-1}$) of *H. verticillata* collected during the period June 16-September 28 and of all other species'

propagules collected on the traps between June 30 and September 28 is shown in Figure 4. Over the entire period, mean biomass flux of *H. verticillata* was significantly greater at site DM ($1.05 \text{ gdw m}^{-2} \text{day}^{-1}$) than at site HC or PB (0.06 and $0.09 \text{ gdw m}^{-2} \text{day}^{-1}$, respectively). One 15-cm-long fragment of *H. verticillata* weighs approximately 0.05 gdw . At site HC, the flux of propagules was $< 0.05 \text{ gdw m}^{-2} \text{day}^{-1}$ until September 28. At sites PB and DM, however, flux rates $> 0.5 \text{ gdw m}^{-2} \text{day}^{-1}$ occurred throughout the season. The mean flux of all other species except *C. demersum*

Table 4

Viability of Plant Propagules Transported to Site DM During the Growing Season, 1994

Species	Plant Form	Date Collected	n	Percent Viable	Percent Viable per Species, All Dates Combined
<i>Vallisneria americana</i>	Whole plant	8/9	14	86	90
	Whole plant	8/30	14	100	--
<i>Myriophyllum spicatum</i>	Fragments	5/23	10	70	70
<i>Najas minor</i>	Fragments	6/7	7	86	76
	Fragments	7/27	10	70	--
<i>Zannichellia palustris</i>	Whole plant	6/7	5	100	100
<i>Chara</i> sp.	Whole plant	6/7	1	100	100
<i>Heteranthera dubia</i>	Fragments	8/9	1	100	100
	Fragments	8/30	7	100	--
<i>Hydrilla verticillata</i>	Fragments	6/16	85	86	70
	Fragments	6/22	125	85	--
	Fragments	7/5	124	61	--
	Fragments	8/9	84	56	--
	Fragments	8/28	29	72	--
	Fragments	9/28	125	61	--

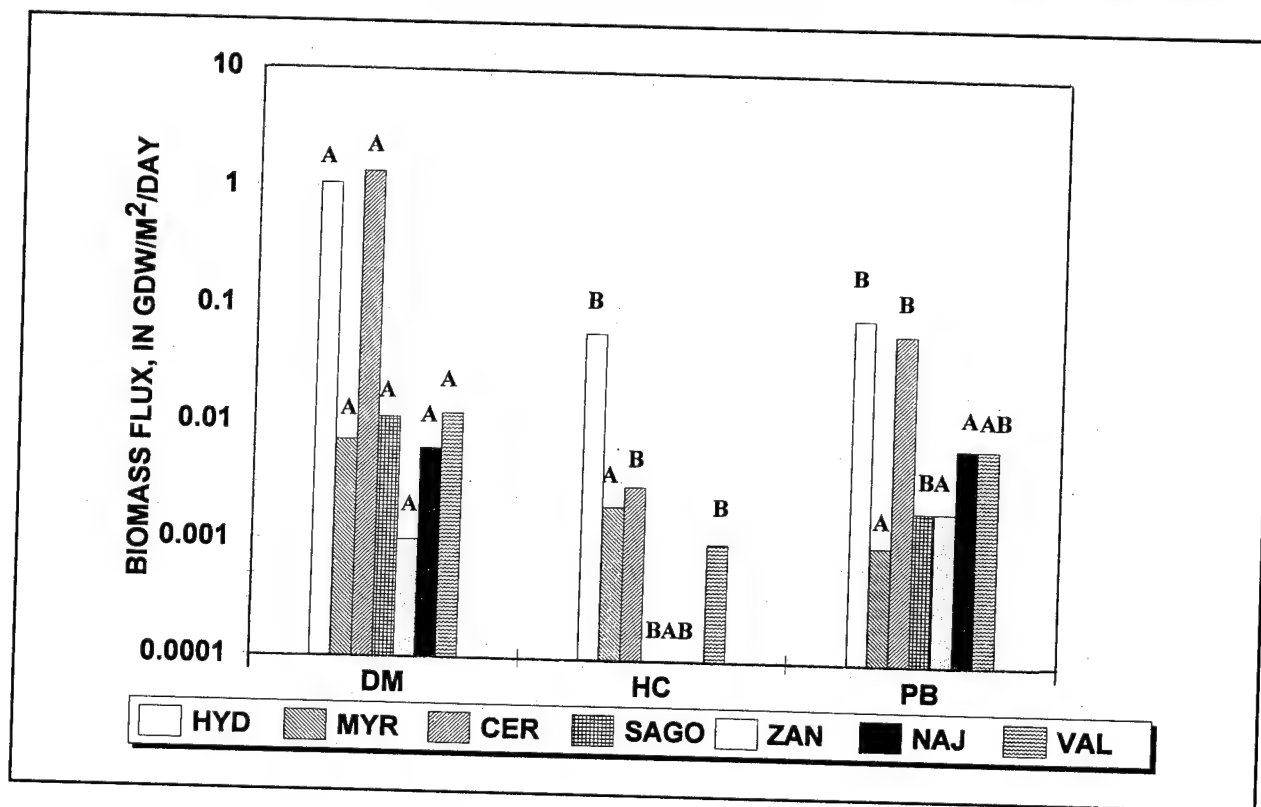


Figure 4. Mean biomass flux (gdw m⁻² d⁻¹) of *H. verticillata* between June 16 and September 28, 1994, and mean biomass flux of all other species between June 30 and September 28, 1994, at unvegetated sites in the tidal Potomac River. (HYD = *Hydrilla verticillata*, MYR = *Myriophyllum spicatum*, CER = *Ceratophyllum demersum*, SAGO = *Potamogeton pectinatus*, ZAN = *Zannichellia palustris*, NAJ = *Najas minor*, VAL = *Vallisneria americana*)

was less than that of *H. verticillata*. Mean fluxes of species other than *H. verticillata* were generally greatest at site DM and least at site HC.

Figure 5 shows mean fluxes of *H. verticillata* and other species at the unvegetated sites by date. At site DM, the fluxes of *H. verticillata* and *C. demersum* were significantly

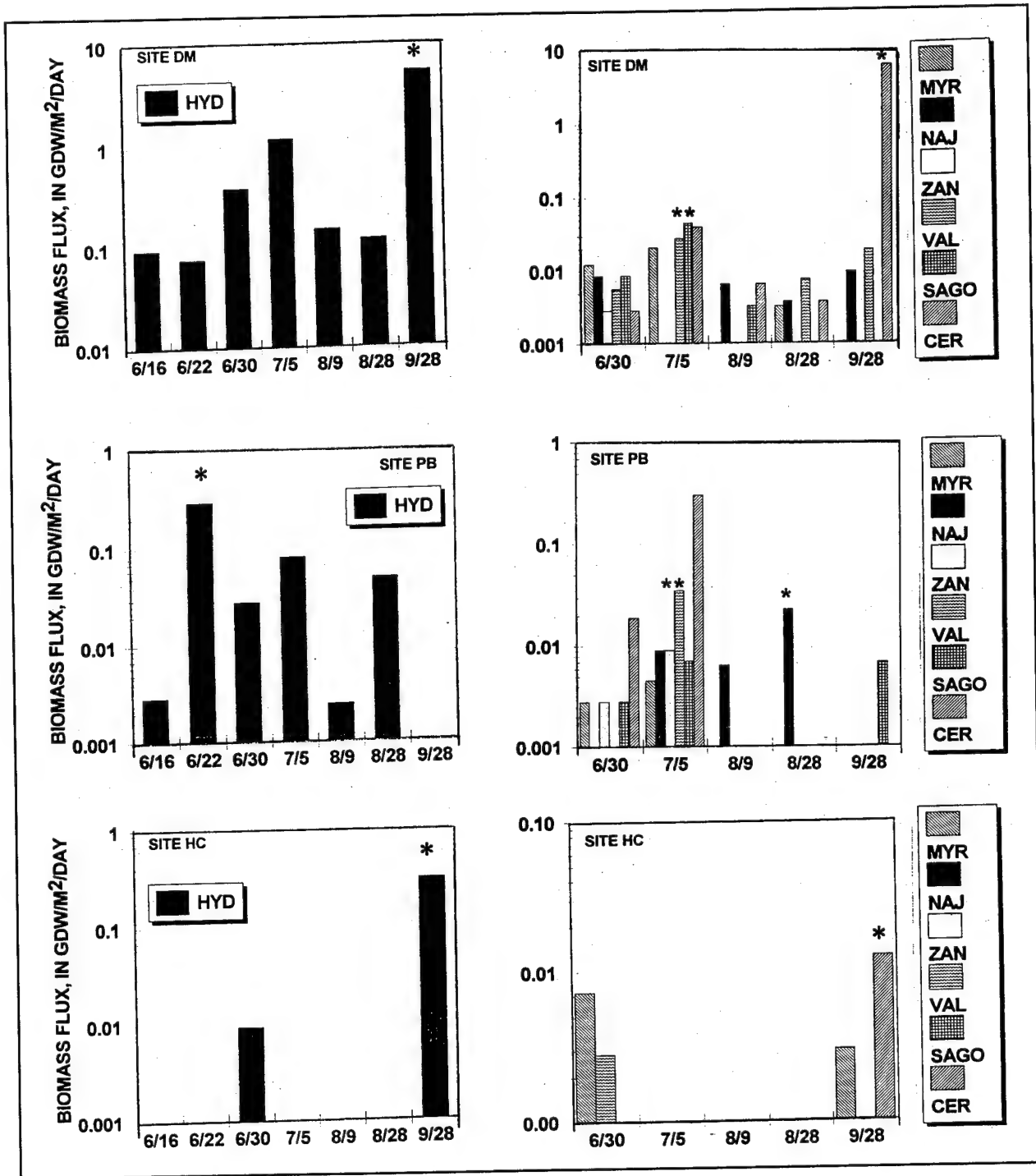


Figure 5. Species biomass flux (gdw m⁻² d⁻¹) by date, at unvegetated sites in the tidal Potomac River.

* indicates species biomass flux significantly different from that on other dates. (HYD = *Hydrilla verticillata*, MYR = *Myriophyllum spicatum*, CER = *Ceratophyllum demersum*, SAGO = *Potamogeton pectinatus*, ZAN = *Zannichellia palustris*, NAJ = *Najas minor*, VAL = *Vallisneria americana*)

greater on September 28, and the fluxes of *V. americana* and *P. pectinatus* were significantly greater on July 5 than on any other date. At site PB, the fluxes of *H. verticillata* were significantly greater on June 22, the fluxes of *V. americana* and *Z. palustris* were significantly greater on July 5, and the flux of *N. minor* was significantly greater on August 28 than any other date. At site HC, the fluxes of *H. verticillata* and *C. demersum* were significantly greater on September 28 than on other dates. Only four of the seven species were ever transported to site HC, whereas all seven species reached sites DM and PB. When all three sites were combined, species flux in the river was significantly greater for *H. verticillata* and *C. demersum* on September 28 and for *M. spicatum*, *V. americana*, and *P. pectinatus* on July 5 than on any other date (Figure 6). Fluxes on nets (not shown) showed the same trends as fluxes on traps; however, fewer species and smaller fluxes were found on the nets.

When flux was analyzed by date and site, there was no obvious relationship among site exposure, average wind direction and speed over the period the traps were out, and biomass of propagules transported to the site; however, wind direction and speed changed frequently during each trapping period. To determine if the wind direction and speed were significant factors in determining flux rate, traps would have to be set out for short periods of time under several different wind conditions. The fact that *H. verticillata* senesces in September was reflected in the significant increase in biomass flux on September 28. Madsen, Eichler, and Boylen (1988) also reported high fragment deposition for *M. spicatum* in Lake George, New York, during senescence in September.

The transect sampling was intended to obtain information on relative biomass and species composition at the six sites to assess the predictive capability of pre-season or post-season propagule data for plant cover during the

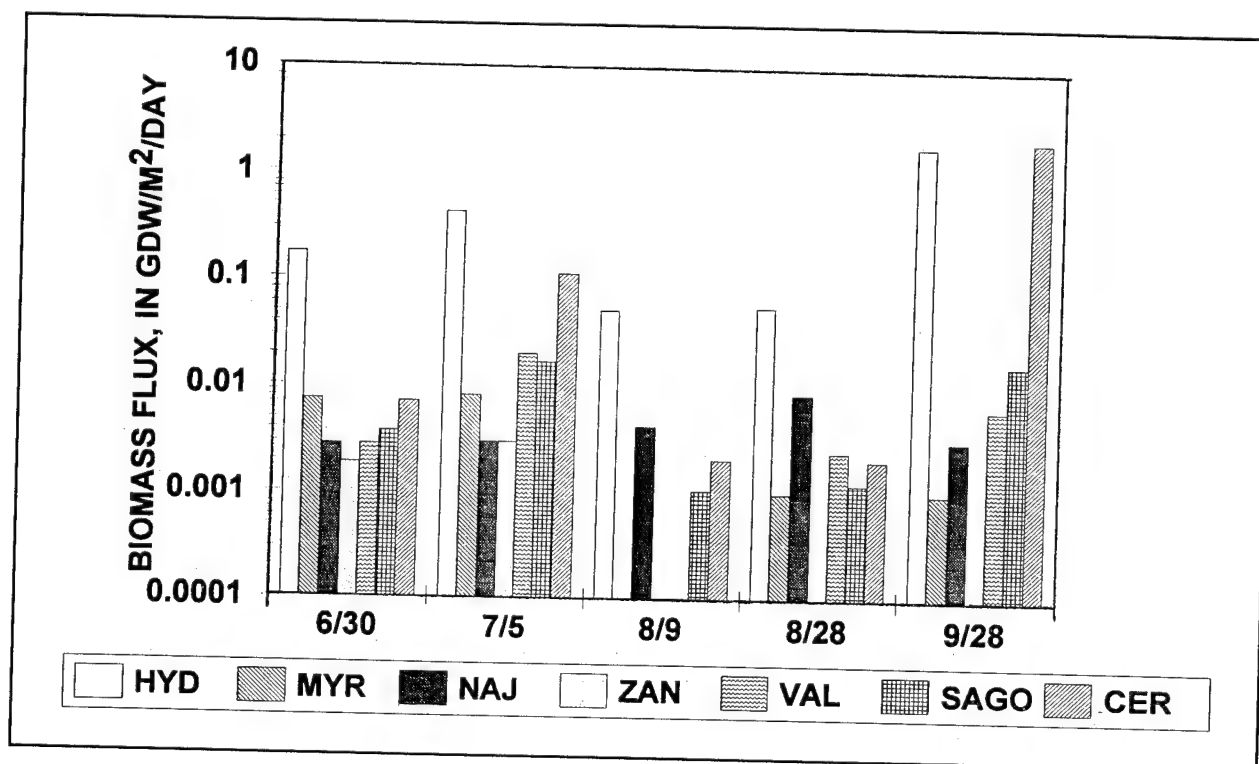


Figure 6. Species biomass flux ($\text{gdw m}^{-2} \text{d}^{-1}$) by date, for all unvegetated sites combined, in the tidal Potomac River. (HYD = *Hydrilla verticillata*, MYR = *Myriophyllum spicatum*, CER = *Ceratophyllum demersum*, SAGO = *Potamogeton pectinatus*, ZAN = *Zannichellia palustris*, NAJ = *Najas minor*, VAL = *Vallisneria americana*)

growing season, and to determine whether biomass was increasing or decreasing. Maximum biomass was found at the three vegetated sites; however, despite its unvegetated status, the largest number of species (five) was found growing at site DM during June (Figure 7, Table 5). Site PB was never vegetated, and site HC had only a small patch of *H. verticillata* nearshore in June and July. Figure 8 compares the biomass of *H. verticillata* with total biomass at all sites on the three sampling dates. There was a trend toward increasing total biomass over time at sites GC and DD and a trend toward decreasing biomass over time at sites GF, DM, and HC. The major species at sites GF and GC was *H. verticillata*; *V. americana* was the dominant species at site DD.

The *H. verticillata* apical stems that had floated in the river for 0, 1, 2, and 4 weeks increased in biomass when planted in the laboratory (McFarland and Barko 1995). The increase was the greatest when plants were left in the river for 2 weeks and least when plants were left in the river for 4 weeks. None of the stems that had floated in the river for 0, 1, 2, and 4 weeks and been placed in

flats on the bottom at site DM grew in the field or were considered viable. Currently, concentrations of plant nutrients in floating fragments are not available.

Figure 9 shows the mean Secchi depth, light attenuation coefficient, chlorophyll-*a* concentration, and total suspended sediment

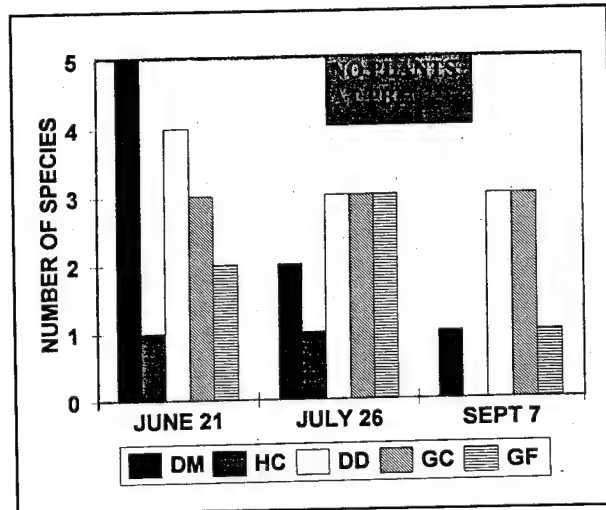


Figure 7. Number of species found at each site in the tidal Potomac River during transect sampling

Table 5
Species Found on Transects at Vegetated and Unvegetated Sites in the Tidal Potomac River in Summer 1994¹

Site	June	July	September
DM	<i>Potamogeton pectinatus</i> <i>Hydrilla verticillata</i> <i>Zannichellia palustris</i> <i>Vallisneria americana</i> <i>Chara</i> sp.	<i>Potamogeton pectinatus</i> <i>Hydrilla verticillata</i>	<i>Hydrilla verticillata</i>
HC	<i>Hydrilla verticillata</i>	<i>Hydrilla verticillata</i>	
DD	<i>Hydrilla verticillata</i> <i>Najas minor</i> <i>Vallisneria americana</i> <i>Chara</i> sp.	<i>Hydrilla verticillata</i> <i>Najas minor</i> <i>Vallisneria americana</i>	<i>Hydrilla verticillata</i> <i>Najas minor</i> <i>Vallisneria americana</i>
GC	<i>Hydrilla verticillata</i> <i>Najas minor</i> <i>Vallisneria americana</i>	<i>Hydrilla verticillata</i> <i>Najas minor</i> <i>Vallisneria americana</i>	<i>Hydrilla verticillata</i> <i>Najas minor</i> <i>Vallisneria americana</i>
GF	<i>Hydrilla verticillata</i> <i>Ceratophyllum demersum</i>	<i>Hydrilla verticillata</i> <i>Zannichellia palustris</i> <i>Vallisneria americana</i>	<i>Hydrilla verticillata</i>

¹ No plants were found on transects at site PB.

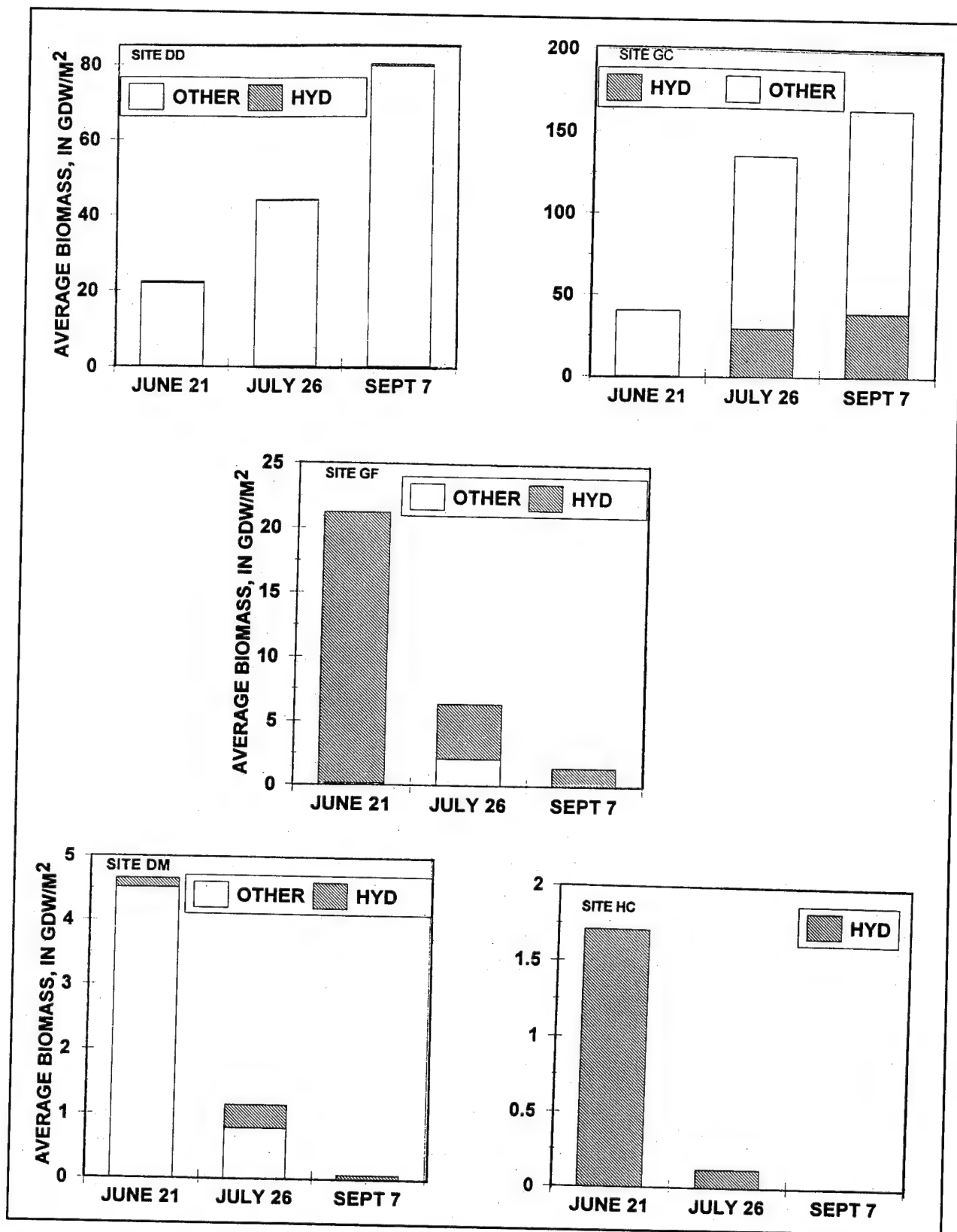


Figure 8. Biomass of *Hydrilla verticillata* and total biomass of all other species combined collected on transects at each site in the tidal Potomac River

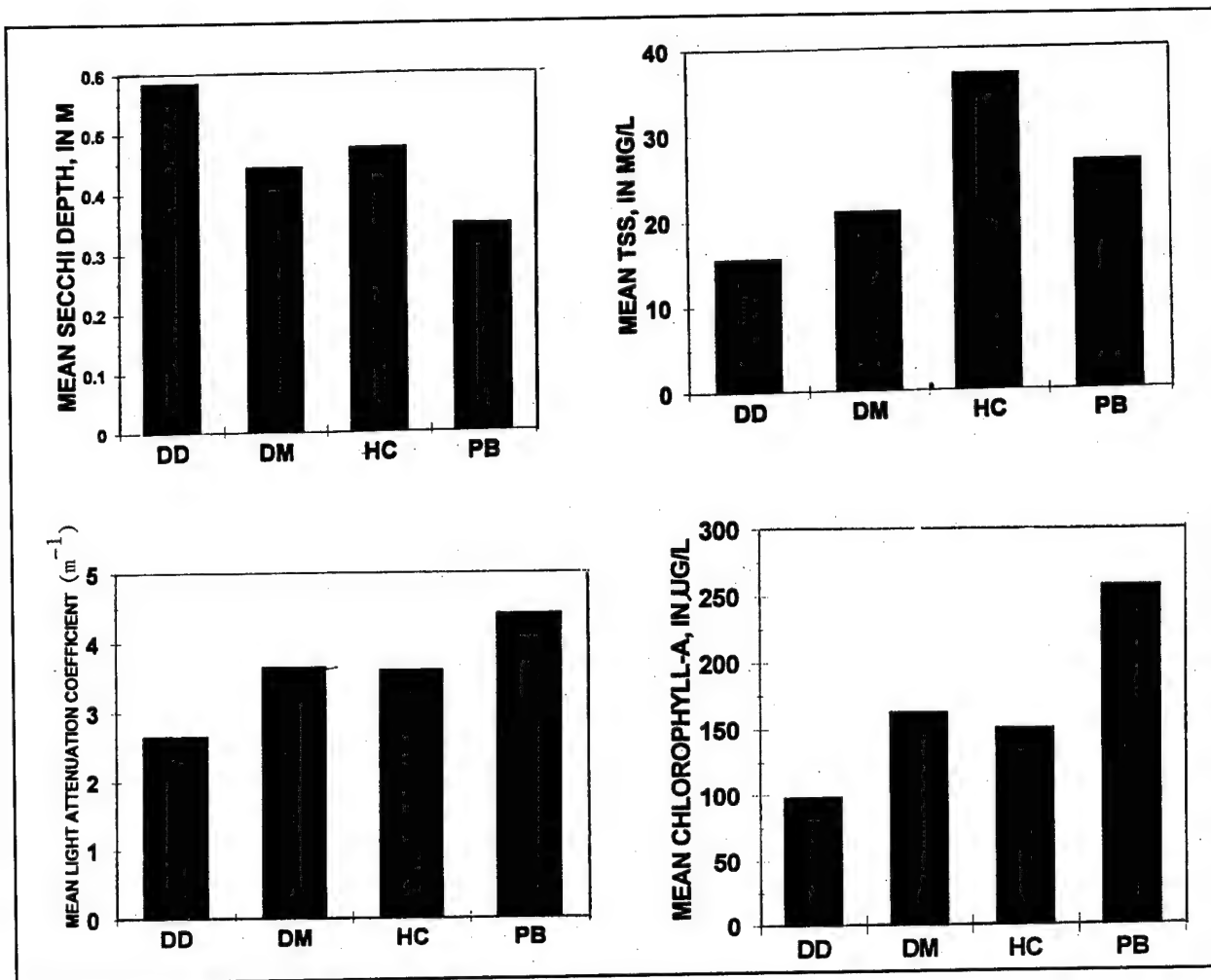


Figure 9. Seasonal (April-September) mean values for Secchi depth, light attenuation coefficient, total suspended sediment (TSS), and chlorophyll-a for sites DD, DM, HC, and PB in the tidal Potomac River

for the period May through September at sites DD, DM, HC, and PB. At site DD, the Secchi depth was higher and the chlorophyll-a concentration, TSS, and light attenuation coefficient were lower than at the three unvegetated sites. Algal growth, as indicated by the extremely high chlorophyll-a concentration, accounted for a majority of the light attenuation.

Discussion

Many factors affect reproduction and survival of macrophytes, as shown in the conceptual model in Figure 10. While the size and number of tubers and their success in germinating are important to recruitment, the plants they produce can survive only if site condi-

tions are adequate. If water clarity is poor or concentrations of sediment nutrients are low in the spring and plants do not survive, fragment flux is the primary new source of vegetation. These propagules, as well, will not survive to reproduce if they do not have enough energy and nutrients to reach the surface. They must also overcome the energy deficit created by leaf loss by herbivory and epiphyte encrustation. Over the winter, some tubers are lost to herbivory and disease, and tubers are washed away from the site or buried by deposited sediment during storms (Carter and Haramis 1980, Rybicki and Carter 1986, Titus and Hoover 1991).

The presence of preseason tubers or turions can help predict the major species

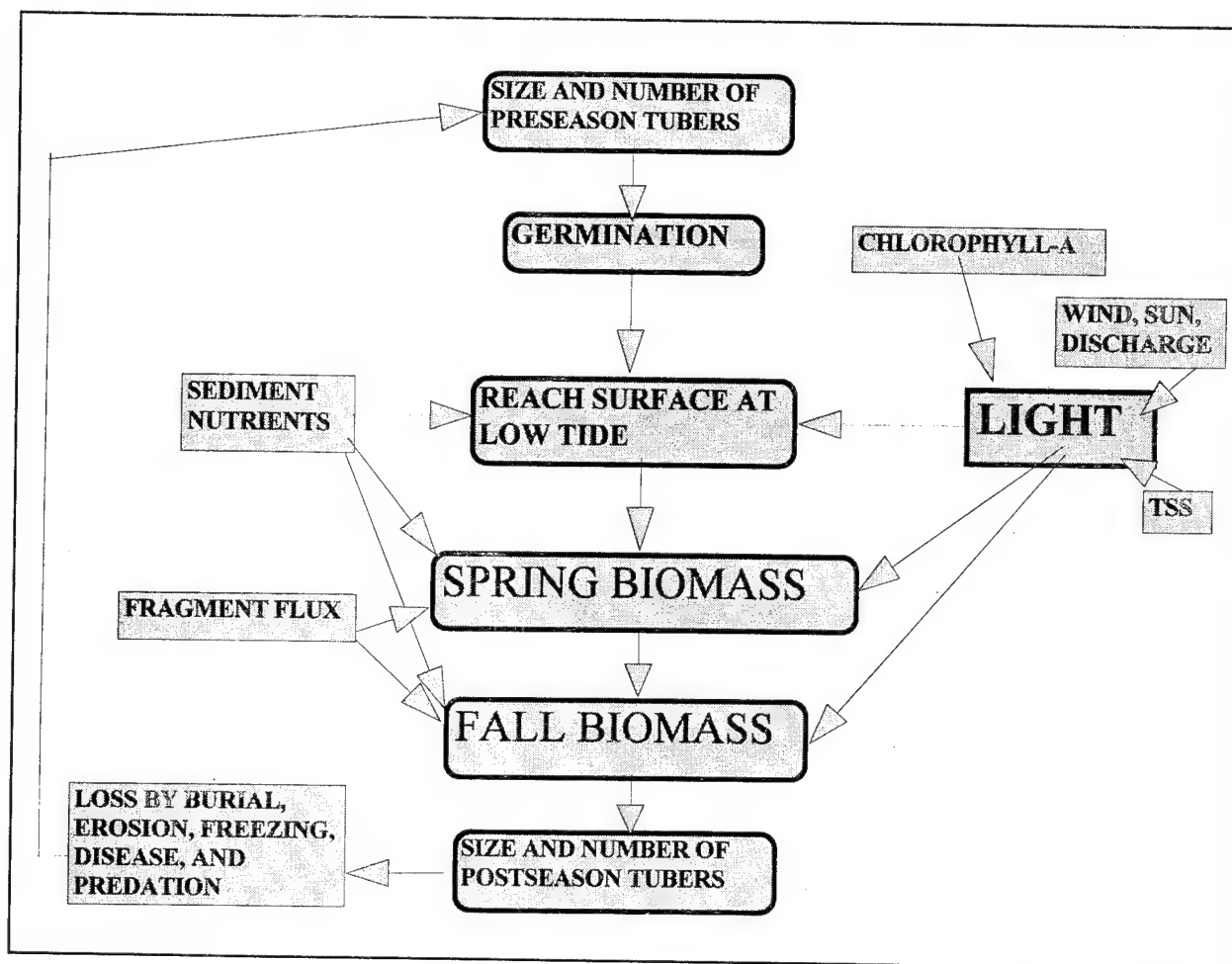


Figure 10. A conceptual model showing the factors involved in reproduction and survival of submersed aquatic vegetation

composition at sites in the tidal Potomac River, but the presence of these propagules does not guarantee high biomass or continued survival of plants at a site. The following predictions are made with this in mind. The biomass of *H. verticillata* declined from 22 gdw m⁻² to near zero at GF by September, and the results of the postseason sampling suggest there will be a very small initial population of *H. verticillata* in 1995 (Figures 2 and 8). Prospects for continued survival of the *V. americana* population at site DD look good because the September biomass of *V. americana* reached about 80 gdw m⁻², and the postseason sampling showed that *V. americana* tubers were present at the site, although the number had decreased substantially compared to the pre-season. Therefore, next year's spring biomass may be diminished. During the pre-season, no tubers or turions were found at the unvegetated

sites. Sites HC and PB remained unvegetated all summer except for a small patch of *H. verticillata* in water <0.5 m deep at site HC, which persisted into July and then disappeared. Re-vegetation from tubers at sites HC and PB in 1995 is unlikely. Site DM had a healthy population of *P. pectinatus* in May and June at the 0.5-m depth. No *P. pectinatus* tubers were found in our pre-season sampling, so most of the 1994 plants may have come from April fragments or seeds. The *P. pectinatus* tubers were set, and seeds were formed on plants growing in water <0.5 m deep at low tide in 1994. It is likely that site DM will again have a good spring biomass of this species.

Pre-season propagules were not an accurate indicator of survival at a site or growth in 1994, nor was propagule flux. Although we assume the mean flux of propagules at the unvegetated

sites was sufficient to cause revegetation if growing conditions had been adequate, transect samplings showed that even the species with the greatest flux were not reestablished. Viability tests showed that most of the fragments found were viable, even after floating in the river for more than a week, so other factors must be responsible for the failure of revegetation.

Poor water clarity could account for lack of growth over the growing season at the unvegetated sites and for a decrease in fall plant biomass at sites DM and GF. Of the four sites where we have water clarity data (DD, DM, HC, and PB), DD had the best water clarity, the greatest biomass throughout the season, and the greatest number of postseason tubers. Previous studies in the Potomac showed that water clarity was an indicator of increase or decrease in vegetation in a reach. A seasonal average Secchi depth of ≥ 0.65 m, TSS ≥ 19 mg L⁻¹, and a chlorophyll-*a* concentration >15.0 μ g L⁻¹ generally caused either no change or a decrease in plant cover from the previous year over the period 1983 to 1989 (Carter et al. 1994). Sites DM, HC, and PB met all three conditions, and site DD met two of the three criteria. Although biomass increased throughout the season at site DD, the number of tubers decreased between the pre- and postseason samplings.

Sediment nutrient deficiencies could be a secondary factor accounting for the failure of unvegetated sites to revegetate and for the decrease in fall biomass. Lower sediment nutrient content results in smaller, shorter plants which may not reach close enough to the surface for survival. Poor concentrations of sediment nutrients do not account for lack of viability of *H. verticillata* apical fragments placed on the bottom of the river at site DM. These fragments deteriorated before they were able to root. Stems appeared unable to produce new growth fast enough, under low light, to compensate for the fragmentation, predation, loss of leaves, and heavy epiphyte load that occurred.

Additional research is needed to quantify the variation in biomass flux seasonally and

from year to year and its effect on revegetation. Factors, such as small patches of plants, water clarity, and sediment nutrients that influence survival and reproductive capacity, should be investigated. If relations could be established among the factors involved with recruitment and revegetation of sites, the information could be used to better understand macrophyte population dynamics and to verify models of submersed aquatic plant growth.

Acknowledgments

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Viability and Growth of Submersed Macrophyte Propagules from the Potomac River: Laboratory Studies

by

Dwilette G. McFarland¹ and John W. Barko¹

Introduction

The submersed aquatic vegetation (SAV) of the freshwater tidal Potomac River has a history of dramatic change in abundance and distribution. Studies by the U.S. Geological Survey and the U.S. Fish and Wildlife Service indicate that following a four-decade absence in the freshwater tidal river (between Chain Bridge and Quantico, VA; Figure 1), SAV reappeared in 1983 in the upper tidal reach and, by 1986, had spread further downstream into the lower tidal reach as well (Paschal et al. 1982; Haramis and Carter 1983; Carter, Paschal, and Bartow 1985; Carter and Rybicki 1986, 1990; U.S. Geological Survey 1994, unpubl.). Three submersed species, *Vallisneria americana* Michx., *Myriophyllum spicatum* L., and *Heteranthera dubia* (Jacq.) MacM., were dominant between 1983 and 1985; however, by 1986, monoecious *Hydrilla verticillata* (L.f.) Caspary, a newly established exotic species (first documented near Dyke Marsh in 1982; Steward et al. 1984) had become most abundant. Since 1986, the SAV in portions of the tidal river has declined, with the upper tidal reach experiencing the most severe losses (U.S. Geological Survey 1994, unpubl.).

To better understand observed changes in the SAV communities, investigations of the tidal river were conducted to determine in situ factors affecting growth. Studies completed thus far generally attribute changes in production and distribution patterns to changes in light availability, and sediment and water quality characteristics (Carter, Paschal, and Bartow 1985; Carter and Rybicki 1985,

1990). However, far less is known of how in situ conditions might influence submersed macrophyte propagation. Information on propagule production and early stages of growth would be useful in predicting the future survival and distribution of SAV in the river.

With these considerations, in March of 1994, the U.S. Geological Survey and the Environmental Laboratory (EL) of the WES began joint investigations of the transport, deposition, and survival of submersed macrophyte propagules in the tidal Potomac River. Laboratory studies conducted at WES focused on monoecious *Hydrilla verticillata*, a prolific species that produces a variety of propagule types (i.e., tubers, turions, fragments, rhizomes, stolons, seeds, etc.). These studies were designed to assess viability of overwintered tubers and turions, and examine growth of fragments exposed to river conditions for different periods of time. Also, to enhance understanding of factors that may limit propagule growth in the river, we compared growth of *Hydrilla* fragments on a reference sediment to sediments from Potomac River sites.

Methods and Materials

Study sites in the Potomac River

Five sampling sites were established in the freshwater tidal Potomac River near Washington, DC (Figure 1). According to recent U.S. Geological Survey data (for June 1993), two of the five sites were vegetated with 40 to 70 percent areal coverage in SAV: (1) Greenway Flats (GF)—covered predominantly with *Myriophyllum* and *Hydrilla*, and (2) the

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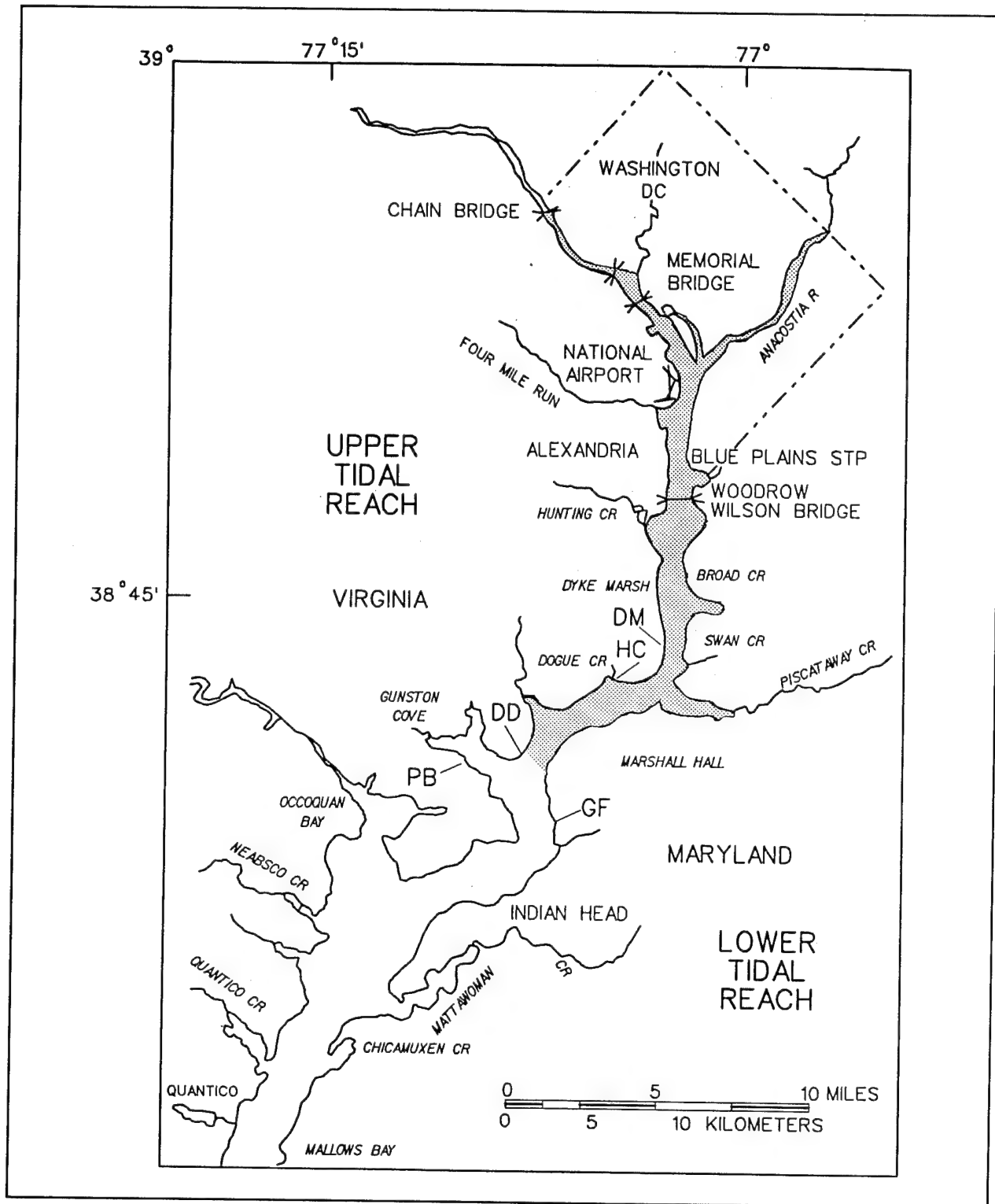


Figure 1. Location of study sites in the Potomac River, 1994

Dempsey Dumpster (DD) site—mainly with *Vallisneria* and *Myriophyllum*; while the remaining three sites were essentially unvegetated: (1) Hunting Creek (HC)—with no SAV, (2) DM-4R (DM)—with only trace amounts

of *Potamogeton pectinatus* L. and *Hydrilla*, and (3) Pohick Bay (PB)—with no SAV. Interestingly, in 1988, the HC and DM sites each had 100 percent areal coverage in *Hydrilla*; the PB site has never supported SAV.

Study 1: Overwintered propagule germination

In March 1994, all five sites were sampled for overwintered tubers and turions of *Hydrilla*. At each site, sampling was conducted at four to five stations along a transect perpendicular to the shoreline; transects were generally terminated at a depth of 2.0 m at low tide (Rybicki and Carter 1995). Tubers and turions in the sampled sediments were removed by sieving through a fine-mesh screen. The collected propagules were refrigerated (at 6 °C) and shipped to WES in coolers within 5 days; there, they were transferred to 6-L battery jars of low alkalinity culture solution (after Smart and Barko 1985) and held in cold storage (at 6-8 °C) for approximately 2-1/2 weeks.

Germination of the propagules was observed under greenhouse conditions in the EL, WES.

Large (1,200-L) white fiberglass tanks were filled approximately 50 cm deep with the same culture solution as indicated above. The solution was maintained at 25 (± 1) °C and aerated with humidified air to enhance mixing and air/water CO₂ exchange. Maximum midday photosynthetically active radiation (PAR) inside the tanks reached moderate levels (350-400 microE m⁻² sec⁻¹) using neutral density shade fabric over the greenhouse roof.

Tubers and turions of *Hydrilla* were planted in sediment collected from Brown's Lake, WES (characterized in Tables 1 and 2); this medium was thoroughly mixed and poured to a depth of 6 cm in small (300-ml) plastic cups. Prewedged propagules were planted singly per cup and observed daily for germination over a period of 2 weeks. Fresh weights determined just prior to planting were later

Table 1
Sediment Physical Characteristics¹

Source	Particle Size		Organic Matter, %	Moisture Content, %	Density mg/L
	Coarse >50 μ diam	Fine $\leq 50\mu$ diam			
Brown's Lake	9.2 c	90.8 a	4.24 a	39.5 a	0.88 b
Pohick Bay	86.7 a	13.3 c	1.81 c	27.3 b	1.23 a
DM-4R	86.7 a	13.3 c	2.20 b	27.7 b	1.17 a
Hunting Creek	67.5 b	32.5 b	2.06 bc	29.0 b	1.17 a

¹ Values are means \pm standard errors based on analysis of three replicate sediment samples. Within each column, means sharing the same letter do not differ at the 5% level of significance (Duncan's Multiple Range Test).

Table 2
Sediment Nutrient Concentrations.¹

Source	Exchangeable Nitrogen, NH ₄ -N, mg/g	Available Phosphorus, PO ₄ -P, mg/g
Brown's Lake	0.069 \pm 0.001 a	0.157 \pm 0.014 a
Pohick Bay	0.034 \pm 0.001 b	0.068 \pm 0.002 c
DM-4R	0.019 \pm 0.001 c	0.110 \pm 0.001 b
Hunting Creek	0.017 \pm 0.000 d	0.065 \pm 0.001 c

¹ Values are means \pm standard errors based on analysis of three replicate sediment samples. Within each column, concentrations sharing the same letter do not differ at the 5% level of significance (Duncan's Multiple Range Test).

used in assessing the viability of propagules in different size groups.

Study 2: Fragment viability

In July 1994, apical fragments 15 cm in length were obtained from a naturally occurring population of *Hydrilla*, 3 km downstream from the PB site (Figure 1). The fragments were divided into two sets: one for EL (see below) and one for field use (described in Rybicki and Carter 1995). Initial samples were taken from each set (i.e., 0 weeks exposure) and the remaining fragments were then floated in exclosures at the DM site (unvegetated, Figure 1) for exposure periods of 1, 2, and 4 weeks.

In the EL, fragments from each exposure group (i.e., 0, 1, 2, and 4 weeks) were subsampled to assess fragment biomass (oven-dried at 80 °C), morphological characteristics (i.e., stem length and number of apices), and nutritional status prior to viability testing. Presently, plant tissues are being analyzed for N and P as described in Barko et al. (1988) and for carbohydrates, i.e., free sugars, starch, and total nonstructural carbohydrate after Nelson (1944), Swank et al. (1982), and Luu and Getsinger (1990).

The viability of fragments was tested under greenhouse conditions similar to those described for Study 1. Twenty fragments per exposure were planted individually in small (300-ml) cups; each cup contained well-mixed sediment collected from Brown's Lake (described in Tables 1 and 2). After 3-1/2 weeks of growth, the plants were clipped at the sediment surface and the shoots measured for morphological responses (as above). Final (i.e., total) biomass was assessed from separate determinations of above- and belowground (oven-dried) plant mass. Viability was evaluated based on changes in total biomass and morphology over the 3-1/2-week growth period.

Study 3: Sediment bioassay

The investigation was conducted in a controlled growth chamber at the EL, WES. This facility was operated to maintain 25 °C and a 14-hr photoperiod at 350 microE m⁻² sec⁻¹.

Hydrilla fragments were grown in 20-L lucite columns (150 cm tall) with removable 3.5-L bases (Figure 2). A detailed description of the environmental chamber and ancillary equipment is presented in Barko and Smart (1980).

Sediments used in the study were collected from Brown's Lake (reference sediment) and from the three unvegetated Potomac River sites: DM, PB, and HC (Figure 1). Each sediment was separately mixed and placed into 1-L plastic containers with a surface area of 95.03 cm²; six replicate containers per sediment were inserted into column bases filled with 2 L of washed silica sand (Figure 2).

The four experimental sediments were analyzed for physical and chemical characteristics. Particle size distribution (texture) was determined using the hydrometer method of Patrick (1958). Sediment moisture content and density were evaluated gravimetrically after oven-drying measured volumes of sediment at 105 °C. Dried samples were placed in a muffle furnace and combusted at 550 °C for estimations of organic matter content from loss of mass following ignition (Allen et al. 1974). Exchangeable ammonium-N was obtained by extraction with 1 M NaCl in a cation exchange procedure modified from Bremner (1965). Available P was obtained using 0.030 N NH₄F and 0.025 N HCl (after Olsen and Sommers 1982). Both N and P concentrations were determined calorimetrically with a Technicon Autoanalyzer II, employing a molybdate method for P and a salicylate method for N (APHA 1985).

Apical cuttings of *Hydrilla* 20 cm in length were obtained from a 6-week-old EL culture stock. This stock was originally established from an earlier collection made near the PB site (Figure 1). Three sprigs of *Hydrilla* were planted per experimental column and submerged in 15 L of culture solution prepared in the same manner as that for the previous two studies. Filtered humidified air was delivered to the columns through plastic air dispersers positioned at the base of the plants.

After 5 weeks of growth, above- and belowground plant structures were harvested,

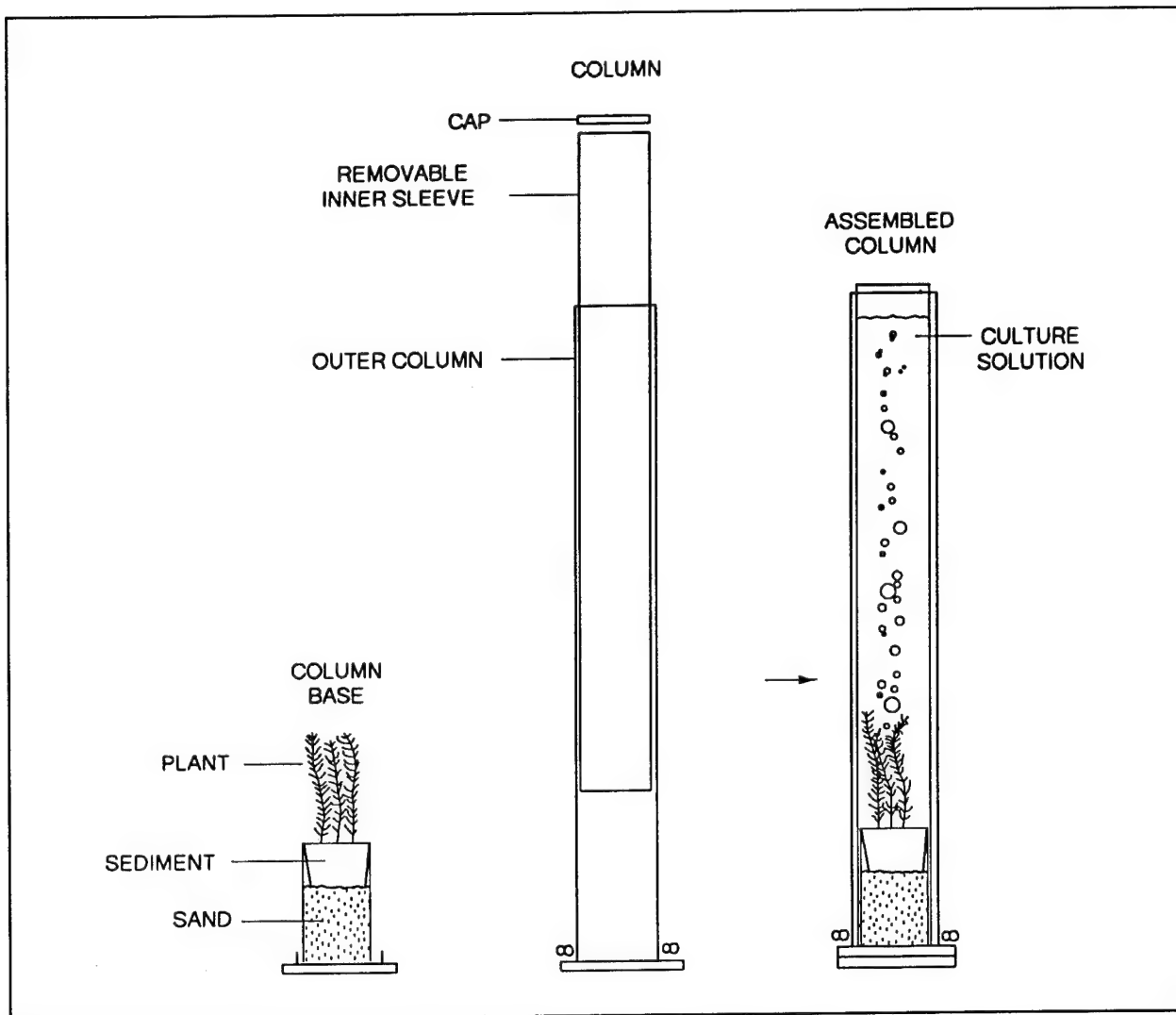


Figure 2. Assembly of lucite columns used in Potomac River sediment bioassay

oven-dried (at 80 °C) and weighed. Total biomass (as the sum of above- and belowground biomass), root-to-shoot ratio, and plant height were used as evaluations of growth.

Data analysis

Data from these studies were analyzed using analysis of variance (ANOVA) and post-ANOVA procedures of the Statistical Analysis System (SAS Institute 1991). Comparisons of means were accomplished using Duncan's Multiple Range Test. Hereafter, statements of statistical significance refer to probability levels of 5 percent or less.

Results and Discussion

Study 1: Germination of overwintered propagules

Nearly all overwintered tubers and turions of *Hydrilla* were found at the GF (vegetated) site. The general absence of these propagules at the other sampling sites was not particularly surprising since, during the previous growing season, three of the five sites (PB, HC, and DM) had been unvegetated and the remaining vegetated site (DD) had been occupied mostly by *Vallisneria*.

The percentage germination of tubers of *Hydrilla* harvested from different depths at the GF site are listed in Table 3. No tubers were found at the shallowest (0.5-m) sampling depth (station 1). Tubers collected from water depths of 1.0 to 1.1 m (stations 2 and 3) occurred in the greatest densities and, under laboratory conditions, exhibited the highest germination percentages. Less than half of the tubers harvested from the 1.5-m sampling depth (station 4) and none of those from the 2.0-m depth (station 5) germinated. The results presented here strongly suggest that environmental conditions at depths of about 1.0 m were more conducive to tuber production than at the other sampling depths. For all stations at the GF site combined, tuber germination ($n = 49$) was approximately 78 percent.

Tubers of *Hydrilla* ranged in fresh weight from 13 to 306 mg. Examination of tuber frequency in different size (fresh weight) categories

revealed a distribution pattern that was slightly skewed to the left (Figure 3). Tubers in the smallest size category (<81 mg) accounted for approximately 30 percent of the total population; however, the percentage germination in these propagules was low (50 percent). Although relatively few tubers accounting for only 12 percent of the population occurred in the largest size category (241-320 mg), the percentage germination was highest (100 percent) in this group.

Eight-one turions of *Hydrilla* were collected from the GF site—most of which were obtained from sampling depths of 1.5 to 2.0 m (stations 4 and 5; Table 3). On the whole, only 60 percent of these propagules germinated successfully under favorable growth conditions, perhaps because many in this population were extremely small. Turions ranged in fresh weight from 4 to 60 mg, and similar to the tuber population, exhibited increased

Table 3
Percentage Germination of Overwintered Tubers and Turions of *Hydrilla* Collected From Different Sampling Stations at Greenway Flats Site, Potomac River

Station	Depth, m	Density ^a , No./m ²	Number Collected, n	Percentage Germination ^b
Tubers				
1	0.5	0.0	0	n.d.
2	1.0	49.5	18	100
3	1.1	65.9	24	75
4	1.5	13.7	5	40
5	2.0	2.2	2	0
Turions				
1	0.5	2.7	1	100
2	1.0	0.0	0	n.d.
3	1.1	0.0	0	n.d.
4	1.5	156.6	57	54
5	2.0	24.8	23	74

^a Density calculations are based on the number of propagules collected using sampling techniques described in Rybicki and Carter (1995).

^b n.d. = no data.

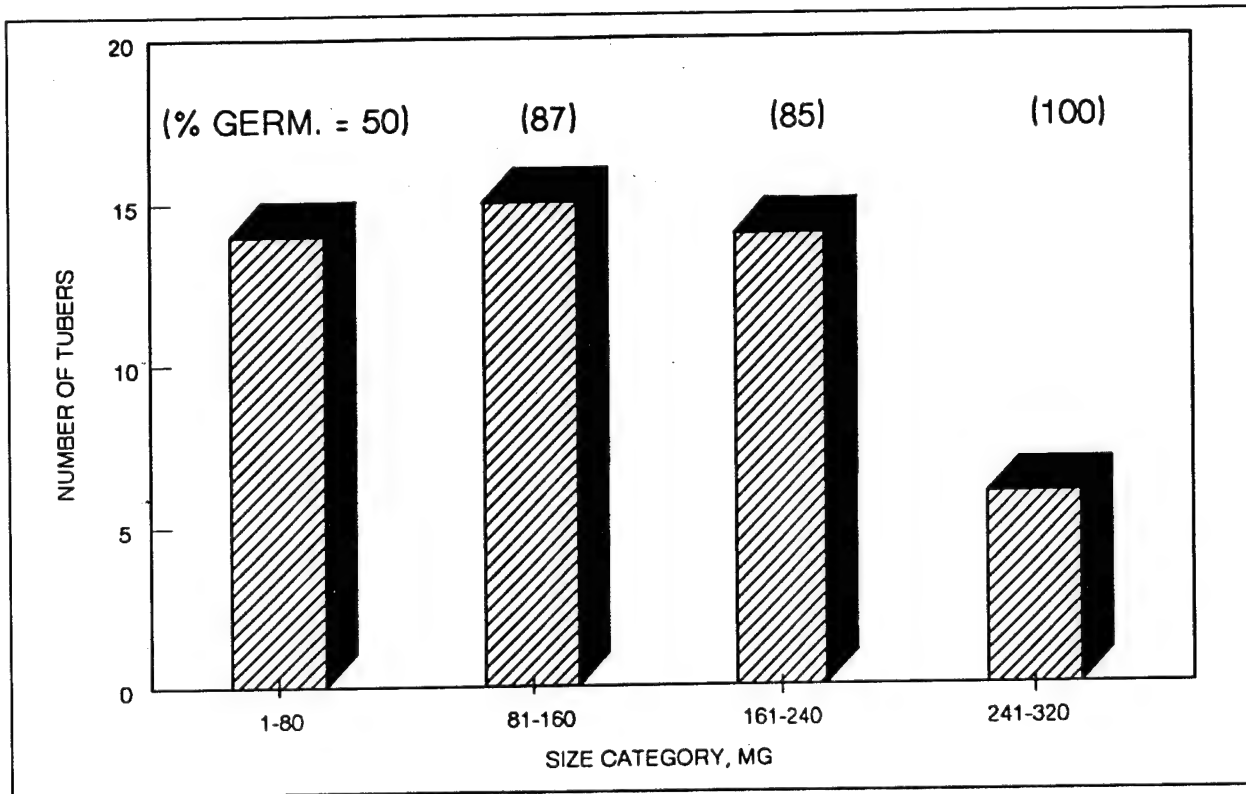


Figure 3. Number of tubers of *Hydrilla* in different size categories by fresh weight collected from the Greenway Flats site. The percentage germination within each size category is given in parentheses

germination with increased propagule size (Figure 4).

Our results provide an initial indication that survival of *Hydrilla* under field conditions could potentially be impacted by propagule mass. At present, influences of tuber and turion mass on success of *Hydrilla* are not well known. However, for *Potamogeton pectinatus* L., germination and initial growth rate have been shown to be positively related to tuber fresh weight (Spencer 1986). Differences in propagule mass effected by changes in environmental conditions (especially temperature and day length), may also influence these processes in *Hydrilla*. Further research of the mass-related vigor of tubers and turions of *Hydrilla* would be useful in predicting recruitment and postgermination success of this species in the field.

In the present study, mean fresh weight per tuber was 146 mg versus 24 mg per turion. Despite anatomical similarities between these

propagule types in *Hydrilla* (Pieterse 1981; Yeo, Falk, and Thurston 1984), the relatively smaller size of turions has been observed in numerous populations of this species (Spencer et al. 1987; McFarland and Barko 1994). Spencer et al. (1987) have proposed that the difference between tuber and turion size may result from different survival strategies. Turions form above ground and thus are more likely to be dispersed by water movements to colonize new habitats. Selective pressures on these propagules would favor, to some extent, a smaller size to aid in dispersal. In contrast, tubers form below ground and, unless there is significant sediment disturbance, are less likely than turions to be transported from place to place. Increases in tuber mass would thus promote propagule stability for reestablishing the population at the occupied site.

Compared to the other four sampling sites (i.e., DD, HC, DM, and PB), GF demonstrated the greatest potential for regrowth of *Hydrilla* from overwintered tubers and turions. However,

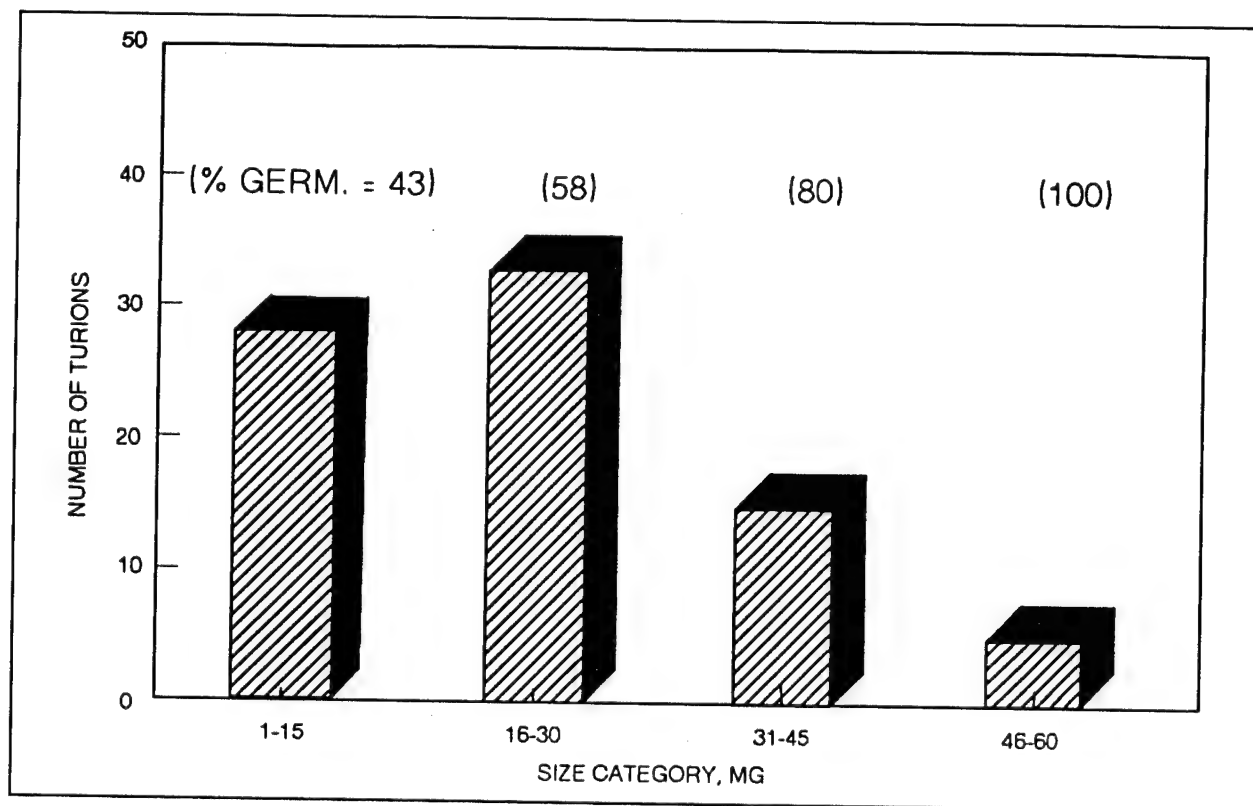


Figure 4. Number of turions of *Hydrilla* in different size categories by fresh weight collected from the Greenway Flats site. The percentage germination within each size category is given in parentheses

findings of this study suggest that the maximum potential for regrowth might not be realized because many of these propagules in our laboratory study failed to grow. In nature, the growth and abundance of *Hydrilla* is potentially influenced by a variety of environmental conditions, e.g., light, temperature, sediment, and water chemistry (Spence 1972, Barko and Smart 1986; Barko, Adams, and Clesceri 1986; Barko, Gunnison, and Carpenter 1991; Barko, Smart, and McFarland 1991; McFarland and Barko 1987; McFarland, Barko, and McCreary 1992; Smart and Barko 1990). From the findings of this study, the viability of tubers and turions could be an important factor as well.

Study 2: Fragment viability

The biomass production in *Hydrilla* was significantly affected by the duration of exposure to river conditions (Figure 5). Fragments that were exposed for 1 week produced as much biomass after planting as did those that

were freshly cut (0 weeks in the river); fragments allowed to float in the river for 4 weeks showed a reduced capacity for growth. The production of biomass was maximized in fragments that were exposed for 2 weeks, probably due to root formation that provided an advantage to these fragments over the others in accessing sediment nutrients. The condition of fragments in the 4-week exposure appeared to be rather poor; there were apparent losses of roots and lower leaves due to grazing, and signs of breakage, yellowing, and general deterioration.

Differences in exposure period also significantly affected shoot height and number of apices in *Hydrilla* (data not presented). Patterns of response essentially paralleled those for biomass production, most notably showing marked reductions in the height and number of apices of fragments floated in the river for 4 weeks.

Analyses of nutrients (N and P) and carbohydrates in the plant tissues are currently

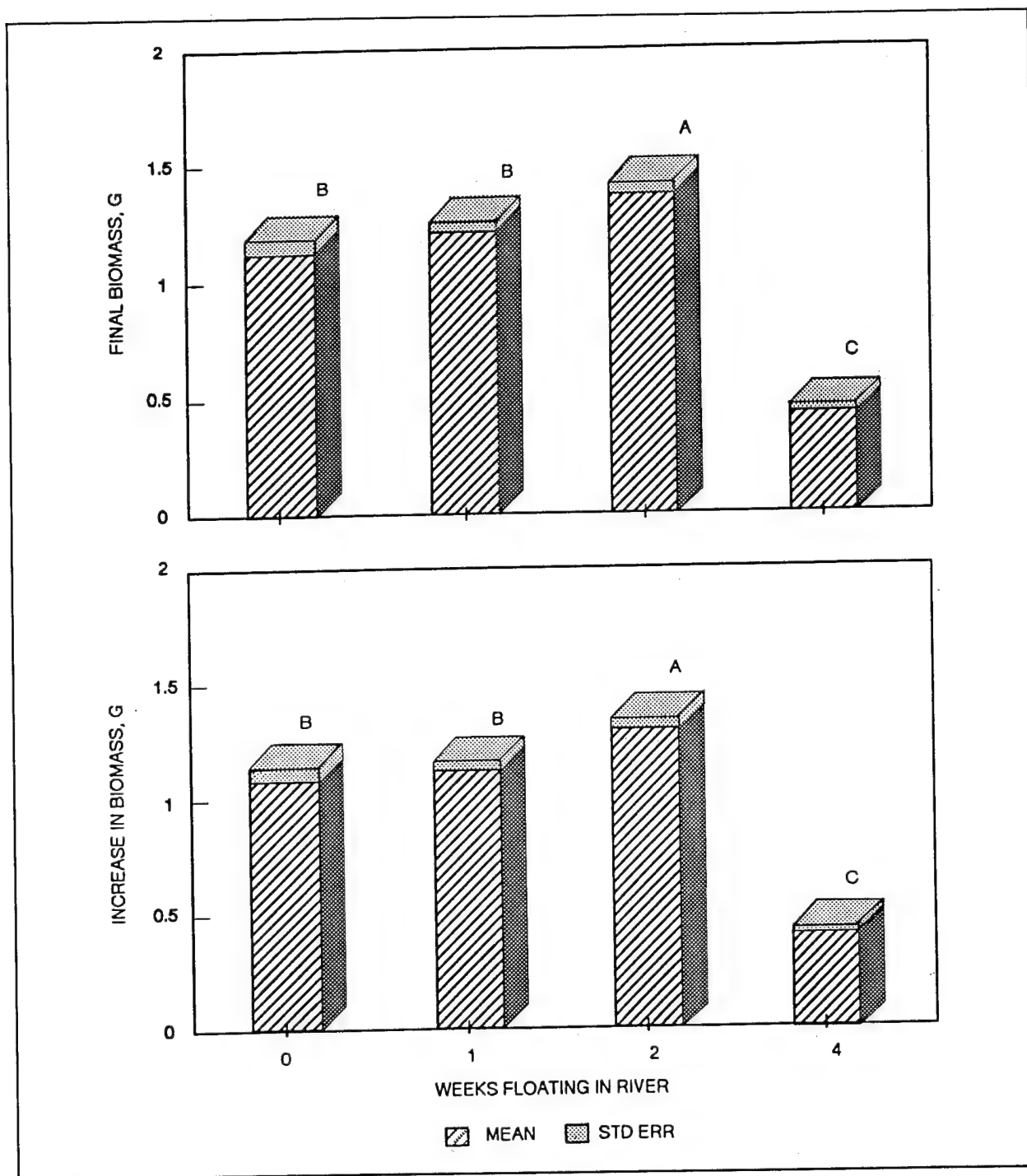


Figure 5. Growth of *Hydrilla* fragments after exposure to river conditions for different periods of time. Values are means and standard errors for final biomass (upper frame) and increase in biomass (lower frame) based on 20 replicate determinations. Within each frame, bars sharing the same letter do not differ significantly from each other. Duncan's Multiple Range Test was used to determine statistical significance at $p < 0.05$

underway. When completed, we hope that the results will provide further insight into the overall vigor of fragments immediately following each exposure.

Study 3: Sediment bioassay

Determinations of sediment compositional characteristics (Table 1) revealed that

Brown's Lake (i.e., BR; reference) sediment was finer in texture than any of the sediments from the unvegetated sites (i.e., HC, PB, and DM). Due to higher sand content (i.e., coarse particle fraction), the PB, HC, and DM sediments each had greater bulk densities, and less moisture than the reference sediment. Organic matter content was greatest for BR, but in all cases, fell within the lower range reported for sediments from North American lakes (Barko and Smart 1986).

Initial concentrations of extractable nutrients differed considerably among the studied sediments, with BR sediment having the highest concentrations of exchangeable N and available P (Table 2). Among sediments from the unvegetated sites, exchangeable N levels were highest in PB sediment, while available P levels were highest in the sediment from DM. HC sediment had the lowest concentrations of exchangeable N and along with PB sediment, the lowest concentrations of available P.

At the end of the 5-week study period, plants on BR sediment were clearly taller and generally possessed greater biomass than on sediments from the Potomac River sites (Figure 6). Reductions in growth were most apparent on HC sediment where total biomass was 25 percent lower and plant height 40 percent lower than on BR sediment. Results of correlation analyses showed that plant height and biomass responses were significantly related to levels of exchangeable N ($r > 0.8$, $p < 0.01$, in both cases) but not available P. Root-to-shoot ratios (though somewhat higher on HC and DM sediments), in general, showed no significant sediment effects.

The results of this study suggest that N availability is likely to play an important role in determining the development of SAV on the studied Potomac River sediments. Similar findings have been reported by others showing N to greatly affect growth of macrophytes both in freshwater and marine systems (Short 1983, 1987; Anderson and Kalff 1986; Duarte and Kalff 1988; McCreary, McFarland, and Barko 1991; McFarland, Barko, and McCreary 1992; Barko and McFar-

land, unpubl. data). Under natural environmental conditions, biological and limnological processes (e.g., sedimentation, mineralization, bioturbation, diffusion, and plant uptake) can contribute to changes in the availability of N and other nutrients in sediment. It is important, therefore, to note that growth of *Hydrilla* relative to nutrient composition of selected sediments was observed here under laboratory (i.e., closed-system) conditions; and thus provides only an indication of potential responses of this species in the field if little or no additional nutrient (especially N) inputs were to occur.

To more accurately assess the potential for nutrient (N) limitation at Potomac River study sites, information regarding nutrient replenishment rates is needed. In aquatic systems, physical disturbances causing the erosion of fine-textured particles and associated nutrients can limit macrophyte growth by promoting nutrient deficiencies (Duarte and Kalff 1988). On the other hand, the accumulation and retention of deposited sediments provide mechanisms of nutrient replenishment and may increase sediment surface area colonizable by SAV (Carpenter 1981; James and Barko 1990). Barko et al. (1988) have suggested that the establishment and vigor of SAV communities depend at least in part on the balance between nutrient gains and losses in littoral sediments. Particular investigative attention should be given to mechanisms affecting this balance at various Potomac River sites.

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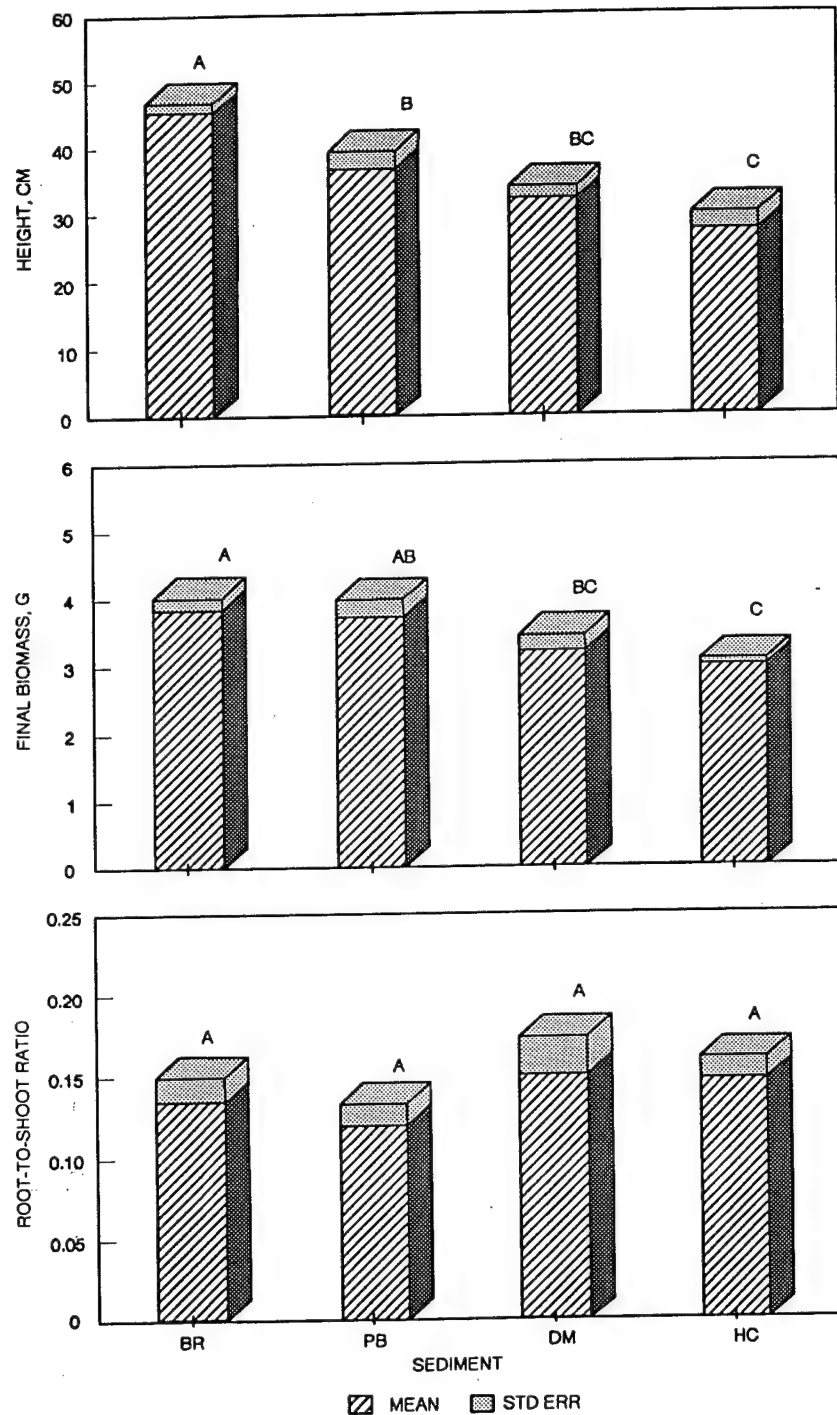


Figure 6. Plant height (uppermost frame), total biomass (middle frame), and root-to-shoot ratios (lowermost frame) in *Hydrilla* grown on sediments from Browns's Lake (BR), and on sediments from three unvegetated sites, Pohick Bay (PB), DM-4R (DM), and Hunting Creek (HC), in the Potomac River. Values are means and standard errors based on six replicate determinations. Within each frame, bars sharing the same letter do not differ significantly from each other. Duncan's Multiple Range Test was used to determine statistical significance at $p < 0.05$

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Preemption: An Important Determinant of Competitive Success

by

R. Michael Smart¹

Introduction

The exotic weedy species hydrilla, *Hydrilla verticillata*, and Eurasian watermilfoil, *Myriophyllum spicatum*, cause problems in many Corps of Engineers reservoirs and waterways. These problem species are highly adapted for exploiting disturbed systems. They exhibit rapid rates of growth and spread, tolerate poor environmental conditions, and grow rapidly to the surface where their leafy shoots form a dense canopy. Since they are so well-equipped to exploit disturbances, these weedy species often grow in large, monospecific (single species) populations.

While a few exotic species cause problems requiring control, most native species do not. These species do not usually form a dense surface canopy and often many species coexist in diverse communities. Native plant communities are desirable because they stabilize sediments, remove excess nutrients from the water, and provide food and shelter for invertebrates and fish.

Our reservoirs and waterways are extreme examples of disturbed systems—they are the result of a near-instantaneous (in ecological time) conversion from terrestrial to aquatic environments. Since these disturbed systems often lack suitable submersed aquatic plants, it is not surprising that many of them are heavily dominated by opportunistic (and usually exotic) weedy species. Although many might view the overwhelming dominance of our reservoirs by exotic, weedy species as evidence of their competitive superiority, these species are actually less competitive than many of our native plants (Smart 1994; Smart and Doyle

1995). The primary advantages that these weedy species have include their early arrival at disturbed sites, their ability to become established rapidly (often under marginal environmental conditions), and (once they establish a foothold) their ability to rapidly spread throughout the system. Other, more desirable species, arriving much later, never have an opportunity to compete with the well-established initial invader. These invasive species are thus *competition avoiders*. They arrive first and exploit the resources of the environment in the absence of competition from other species. This *preemptive growth strategy* accounts for the widespread distribution and dominance of exotic species such as hydrilla and Eurasian watermilfoil (Smart and Doyle 1995).

If we could eliminate the ability of exotic weedy species to preempt resources, many competitive native species should be favored, and we might be able to manage our reservoir and waterway ecosystems to foster development of diverse native plant communities. The effectiveness of long-term aquatic plant management would undoubtedly be improved by promoting the development of communities of competitive native species to resist re-invasion by weedy species. This approach not only offers the potential of prolonging the effectiveness of control operations, but also improves aquatic habitat, possibly at a lower overall cost of management.

The primary objective of research in this work unit is to evaluate the use of communities of competitive and desirable native species to prevent or delay the recurrence of weedy, exotic species. In this article I review recent

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research on preemption as an important factor in affecting the outcome of competition between exotic and native species and briefly describe the status of current and future research in this area.

Small-scale Studies

Previous research in this work unit (reviewed in Smart 1994) identified several native species with desirable characteristics and some competitive abilities. These include: American pondweed (*Potamogeton nodosus*), water star grass (*Heteranthera dubia*), and wild celery or vallisneria (*Vallisneria spiralis*). Each of these species should be examined further to evaluate the beneficial effects of a period of preemption. In this article I present preliminary results of such an investigation. The following greenhouse tank study considered the importance of preemption in affecting the outcome of competition between vallisneria and hydrilla.

Methods

The study involved growing vallisneria and hydrilla alone (without competition) and in mixtures. The experiment was conducted in 1,200-L, temperature-controlled ($25 \pm 1^\circ\text{C}$) fiberglass tanks in a greenhouse facility at the LAERF (Smart and Decell 1994). Plants were grown at a high level of inorganic carbon supply (aeration with $10\times$ ambient CO_2), on sediment of high fertility (fresh Lewisville pond sediment), at moderately high light levels (approximately 50-60 percent of full sunlight). These conditions are quite similar to those used in earlier greenhouse competition studies between vallisneria and hydrilla (Smart 1992, 1993; Smart, Barko, and McFarland 1994).

Each tank contained fifty-four 1-L plastic pots. At the beginning of the experiment, all of the pots in a given tank (treatment) were planted with 2 vallisneria plants, 2 hydrilla apical shoots (15 cm length), or 1 propagule of each species. Another treatment was planted with 1 vallisneria at the beginning of the experiment and the hydrilla was not planted until 4 weeks later to allow for a period of preemption by the vallisneria. After a

9-week period of growth, 10 replicate containers from each treatment were harvested for biomass determination.

At the end of the 9-week experiment, all remaining pots of plants were removed to a LAERF pond to undergo winter conditions. In the spring of the following year, the containers of vallisneria plants were collected from the pond, placed in a greenhouse tank, and a portion of these were planted with hydrilla to determine if vallisneria gained any additional competitive advantage from a longer period of preemption. Conditions during this phase of the experiment were similar to those during the preceding 9-week period of growth.

Results

Results of the two-phase greenhouse experiment are summarized in Figure 1. *Vallisneria* transplants are slow to establish (Doyle and Smart 1993), and, lacking a period of preemption, *vallisneria* was suppressed by hydrilla (Smart 1993). However, when given a 1-month period in which to become established, *vallisneria* was much more competitive and suppressed the growth of the later arriving hydrilla. Extending the period of preemption through the late fall and winter increased *vallisneria*'s competitive advantage even further.

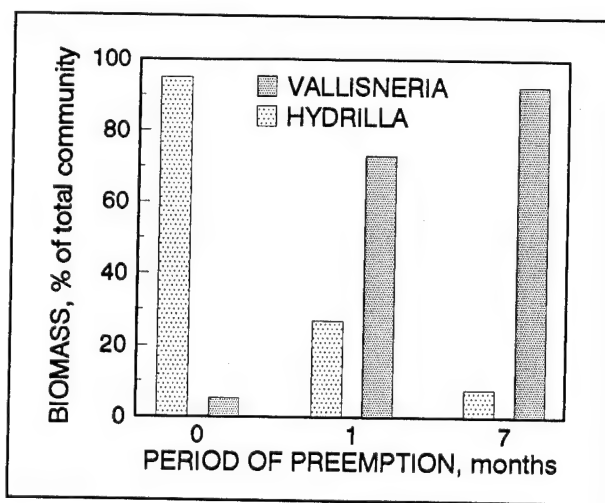


Figure 1. Total biomass, expressed as a percent of the total community biomass, of vallisneria and hydrilla grown together in pot mixtures in a two-phase greenhouse tank experiment. Values are means of 10 replications

Although results of a small-scale, short-term study such as this must be extrapolated with caution, these results do support earlier claims that established *vallisneria* populations should be able to resist invasion by *hydrilla* (Smart 1993). The level of growth that *hydrilla* achieved in competitive mixture with 7-month-old *vallisneria* was minimal, and even this meager growth depended on intentional planting of healthy apical tips into the containers of *vallisneria*. Additional studies, conducted under natural conditions and for longer periods, are needed to substantiate these preliminary results and also to identify longer term (or larger scale) processes, natural events, or larger scale phenomena that may affect competitive outcome. These higher level considerations include, for example, seasonal (phenological) changes in physiology and growth, the effects of a winter period, and the influence of other components of the ecosystem including herbivores as well as competing autotrophs such as algae.

Intermediate-scale Studies

During FY92 and FY93 we conducted an intermediate-scale study of competition over two growing seasons using potted plants placed in a pond environment. This study was designed to examine the importance of seasonal events and phenological stage of development on the mechanics of competition. Specifically we were interested in the relative competitive advantages of different physiological mechanisms of perennation (overwintering), such as the onset and duration of the dormant period and the ability to sequester reserves in dormant tissues. This study also included an examination of the competitive benefits that various periods of preemption conferred on the native plants *vallisneria* and American pondweed.

Methods

In this study 5-L containers were filled with a fertile soil mixture and arranged in groups in a geotextile- and gravel-lined pond. The containers were grouped into "populations"

of a single species and planted in May 1992. Two native species (American pondweed and *vallisneria*) and two exotic species (*hydrilla* and Eurasian watermilfoil) were included in the study although only the *vallisneria* results are considered here. *Vallisneria* transplants and *hydrilla* apical tips served as propagules. Competing *hydrilla* plants were introduced to some of the populations of *vallisneria* by planting apical tips into each of the containers. Three "invasions" by *hydrilla* occurred at 0, 12, and 24 weeks after planting. These times were chosen to evaluate the effects of preemption on the outcome of competition between these two species. Similar monospecific populations of *hydrilla* were also initiated at each of the three planting dates. These are referred to as "cohort 1, 2, or 3." At intervals over the next two growing seasons (16 months), six replicate containers (12 for the final sample date) were harvested from the monospecific populations and the mixed communities. Plants were separated by species, dried to constant mass, and weighed to determine biomass.

Results

Vallisneria transplants are slow to establish (Doyle and Smart 1993) and very little growth of this species occurred during the first 12 weeks of the study (Figure 2A). However, by late August of the first season, *vallisneria* populations had accrued a biomass of approximately 20 g dry mass per pot and held this level throughout the winter. Additional growth of this perennial species occurred during the subsequent spring and the plants accumulated additional biomass during the second growing season.

Hydrilla, planted in May, exhibited rapid rates of growth after a much shorter lag period of 4 weeks (Figure 2B). Populations of *hydrilla* reached biomass levels of 40 to 50 g dry mass per pot during the first season of growth, whether planted in early May (cohort 1), or 12 weeks later in late July (cohort 2). Both *hydrilla* populations maintained these high biomass levels throughout the winter and early spring, but then both declined in the second summer.

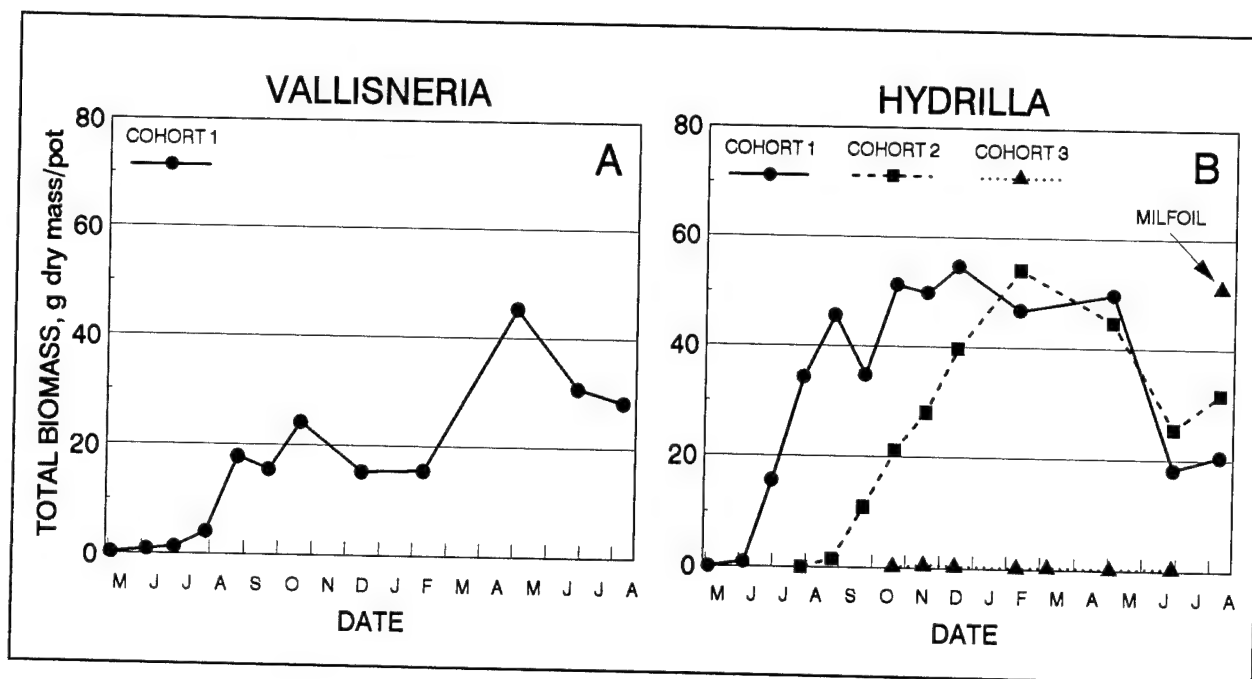


Figure 2. Temporal changes in total biomass of vallisneria (A) and hydrilla cohorts (B) over the course of the pond experiment. Values are means of 6 replications (12 replications for the final sample date). Total biomass of Eurasian watermilfoil (which replaced hydrilla in cohort 3) is plotted for the final sample date (B)

The growth of hydrilla was curtailed at the lower temperatures occurring immediately after the planting of the third cohort in October. Hydrilla planted at this time was unable to accrue sufficient biomass to prevent invasion by Eurasian watermilfoil, which by this time had begun to spread throughout the pond. By the spring of the second season, virtually all of the cohort 3 hydrilla containers had been invaded by Eurasian watermilfoil. Since the presence of Eurasian watermilfoil was noted, but not always quantitatively analyzed, Eurasian watermilfoil data are presented for only the final harvest in Figure 2B. The unplanned Eurasian watermilfoil population accrued 50 g dry mass per pot in its first season, remarkably similar to values measured for the hydrilla populations.

Results of the competition treatments (mixtures) are shown for the final sample date in Figure 3A, B, and C. Biomass of the uninvaded vallisneria population is shown for reference in Figure 3D along with indicators showing the timing of the three hydrilla invasions in relation to the biomass level of the recipient vallisneria populations.

Hydrilla completely eliminated vallisneria from the mixture when the two species were planted together at the same time (Figure 3A). Giving vallisneria a 12-week period of pre-emption did not alter the results appreciably, and these mixtures were also strongly dominated by hydrilla (Figure 3B). Since vallisneria had achieved very little growth prior to the first two hydrilla invasions, these results are not very surprising. However, by the time of the third invasion, in October, vallisneria had achieved considerable growth, and these well-established plants resisted invasion by both hydrilla (which was intentionally planted) and Eurasian watermilfoil (which completely surrounded the vallisneria-hydrilla mixture). Even though the biomass of vallisneria in the mixture was somewhat suppressed relative to that of the vallisneria monoculture these plants were very effective at preventing the establishment and growth of invaders.

These results indicate that, given ample time to establish, vallisneria populations are resistant to invasion by exotic species.

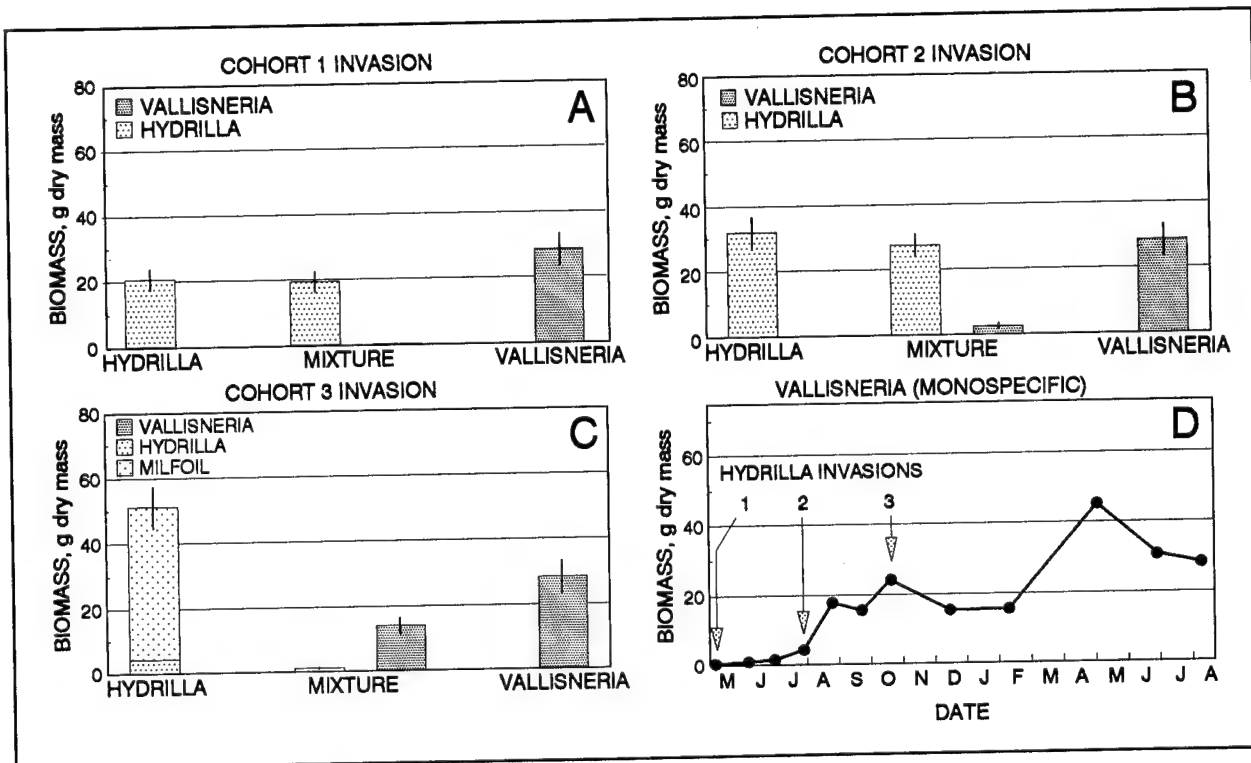


Figure 3. Total biomass of vallisneria and hydrilla achieved in monoculture populations and in mixtures at the conclusion of the pond experiment (A through C). The timing of hydrilla invasions is shown for reference in relation to vallisneria biomass in monoculture (D). Values in A through C are means of 12 replications. Total biomass of Eurasian watermilfoil (which replaced hydrilla in the cohort 3 invasion) is also plotted in C

Large-scale Studies

The experimental objectives of the large-scale study initiated in FY94 were to evaluate a method for establishing founder populations of vallisneria in a reservoir and to determine the ability of these newly established populations to resist invasion by exotics. North Lake, a power supply and recreation lake operated by Texas Electric Utilities and the city of Irving, TX, was selected for the study. This small reservoir was selected due to its proximity to LAERF, its clear, low nutrient waters, and its plant community (which included several native pioneer species). This lake had also been previously treated with Sonar to control hydrilla, and although no hydrilla was found during several surveys of the lake, there was a very high probability that dormant hydrilla tubers remained in the sediment.

Vallisneria was transplanted into nine sites in the lake and successfully established in

most of these. Hydrilla began to regrow throughout the lake during the summer of 1994. The ability of the newly established vallisneria populations to resist the recovering hydrilla will be assessed during FY95.

Future Research

Small-scale studies

Since preemption of sediment nutrient resources seems to be a key factor allowing native species to successfully resist invasion by exotics, short-term studies should examine the importance of nutrient uptake (particularly ammonium or nitrate nitrogen) from the water column. Where nitrogen is readily available from the water column, preemption of sediment nitrogen resources may not be sufficient to ensure persistence of native species in the presence of invading exotic species. Under conditions of greater water column nutrient availability, exotic species such as hydrilla

(which has a greater proportion of shoot biomass) may be favored over more nutritionally conservative, and heavily rooted, species like *vallisneria*. A small-scale study has been initiated to evaluate the importance of water column nutrient availability in affecting the outcome of competition between an exotic weedy species (*hydrilla*) and a competitive native species (*vallisneria*).

Intermediate-scale studies

A pond study of competition between water star grass and *hydrilla* was initiated in FY94. This study also examines the role of preemption in affecting the outcome of competition between these two species in a pond environment.

Large-scale studies

Founder populations of *vallisneria* in North Lake will be monitored during FY95. Additional founder populations of American pondweed and water star grass have been established in Lewisville Lake, a Corps reservoir adjacent to LAERF. These plots will also be monitored during FY95.

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Competitive Interactions of Native Plants with Nuisance Species in Guntersville Reservoir, Alabama

by

Robert D. Doyle¹ and R. Michael Smart¹

Introduction

Established native aquatic plants can provide numerous benefits to a reservoir system such as providing quality habitat for fish and waterfowl and improving water quality. In addition, these beneficial plants may confer an additional advantage of protecting the water body from invasion of nuisance exotic species such as hydrilla (*Hydrilla verticillata*) or Eurasian watermilfoil (*Myriophyllum spicatum*). Previous research at the greenhouse and pond scale (Smart, Barko, and McFarland 1994, Smart 1992, 1993) has demonstrated that established populations of vallisneria (*Vallisneria spiralis*), a beneficial native submersed plant, can effectively resist invasion by aggressive exotics. However, this competitive advantage is realized only when vallisneria is allowed a preemptive period for establishment prior to being subjected to an invasion.

The Guntersville Joint Agency Program has provided the opportunity to extend this plant competition research to the *field trial* level. While numerous competitive interactions have been examined, each has developed to test the central hypothesis that established populations of beneficial native plants would be able to withstand competitive pressure from the nuisance species while mitigating the impact of the problem plants. Previous work in Guntersville Reservoir under this work unit has (1) developed methods for establishment of some native plant species (Doyle and Smart 1993a; Smart et al. 1993), (2) examined the ability of small plots of vallisneria and American lotus (*Nelumbo lutea*) previously established in Guntersville Reservoir to withstand reinvasion by Eurasian

watermilfoil (Doyle and Smart 1994), (3) studied the competitive abilities of several native macrophyte species in shallow areas of the reservoir dominated by the nuisance, mat-forming, cyanobacteria lyngbya (*Lyngbya wollei*) (Doyle and Smart 1993b), and (4) further investigated the interactions of pickerelweed (*Pontederia chordata*) and lyngbya when grown together in containers (Doyle and Smart 1994). In this report we examine the second year of field data for three competitive interactions between beneficial native plants and nuisance species in Guntersville Reservoir. The competitive interactions examined and the locations within Guntersville Reservoir where the field plots were located are:

- (1) Vallisneria:Eurasian watermilfoil (Chisenhall Embayment)
- (2) American lotus:Eurasian watermilfoil (Osa-Win-Tha Cove)
- (3) Pickerelweed:lyngbya (Waterfront Cove)

Vallisneria: Eurasian Watermilfoil Competition

Methods

Detailed information on the methods utilized to establish the native plants is provided elsewhere (Doyle and Smart 1993a) and will be only briefly summarized here. Small (1.5-by 1.5-m) plots of vallisneria were initially planted within a large protective enclosure in Chisenhall Embayment in the spring of 1991. The original plantings were made with dormant winterbuds of vallisneria collected from

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the Holston River, TN. However, despite the protective enclosure, the plots were subject to continuous herbivory by both red-eared pond sliders and muskrat. Even though the plots were replanted several times with tubers from the Holston or transplants from the LAERF, it was not until secondary fences of wire mesh were placed around each small plot and an aggressive muskrat trapping program was in place that the vallisneria began to establish. The surviving vallisneria plots were developing moderate aboveground biomass in 1993 and 1994 (Figure 1).

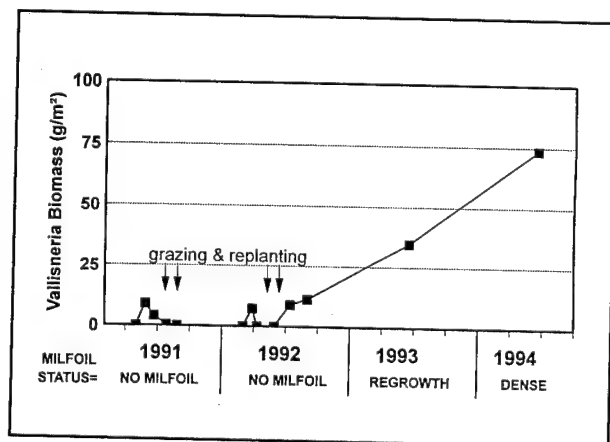


Figure 1. Mass of vallisneria within small plots at the Chisenhall Embayment on Guntersville Reservoir during the experiment period. Also noted is the status of Eurasian watermilfoil over the same time period

Although this embayment had a history of dense infestation, there was virtually no Eurasian watermilfoil present during the initial establishment period (1991-1992). However, by 1993 there was noticeable Eurasian watermilfoil regrowth, especially within the enclosure and by 1994 it had again reached nuisance proportions within the entire embayment (Figure 1). This regrowth pattern of Eurasian watermilfoil allowed us the opportunity to examine the ability of the moderately established plots of vallisneria to resist reinvasion as Eurasian watermilfoil returned to the embayment.

Harvests were conducted in August of 1993 and again in August of 1994 to quantify the development of both vallisneria and Eurasian watermilfoil during the approximate time

of maximal seasonal biomass. In August 1993 a single 0.25-m² quadrat was harvested from within each of the nine vallisneria plots. Of these, only four plots appeared to be moderately established with vallisneria, while the remaining had few surviving vallisneria plants. In addition, six unplanted reference plots within the enclosure were harvested to quantify Eurasian watermilfoil growth in the absence of competition from planted native species. In August of 1994 triplicate 0.25-m² quadrats were collected from the single vallisneria plot which had survived. This plot was growing vigorously beyond the original plot. For comparison, six harvests were also made of adjacent, unplanted areas.

Results and discussion

Eurasian watermilfoil regrowth was significantly lower in plots where vallisneria was established than in adjacent, control plots (Figure 2). The five plots where vallisneria was planted, but where it had not established, had high Eurasian watermilfoil comparable to the unplanted controls. In 1993, when Eurasian watermilfoil was just beginning to regrow, established vallisneria plots contained

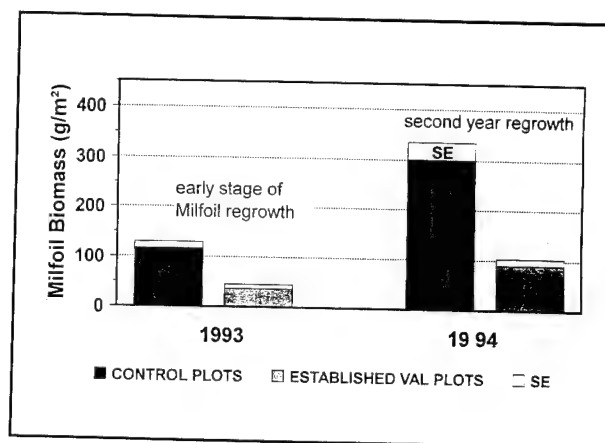


Figure 2. Eurasian watermilfoil shoot mass in small plots of established vallisneria or in adjacent, unvegetated control plots. In 1993 Eurasian watermilfoil was just beginning to regrow within the embayment; in 1994 it had again achieved nuisance biomass levels. In both years the mass was significantly lower ($p < 0.05$) in the vallisneria plots than in the control plots

only about one third of the biomass of the exotic weed than the control plots. This same proportion held in 1994 as the Eurasian watermilfoil population in unplanted plots again achieved nuisance levels (ca. 300 g/m²).

The loss of three moderately established plots of *vallisneria* between 1993 and 1994 is cause for concern. Clearly, in areas where herbivory can be expected, small plots of natives simply do not provide the long-term resiliency needed.

Despite the complicating factor of herbivory, these results support the data generated on the competitive ability of *vallisneria* in greenhouse and pond-scale research. In greenhouse studies, when *vallisneria* is well established in containers and allowed to preempt available resources, it is able to prevent the development of nuisance levels of hydrilla (Smart, Barko, and McFarland 1994; Smart 1992, 1993). Under those conditions, the outcome of 8 weeks competitive growth favors *vallisneria* by a large margin (93 percent *vallisneria*:7 percent hydrilla). However, when planted at the same time, which allows no preemptive establishment of *vallisneria*, hydrilla completely dominates the competitive interaction (5 percent *vallisneria*:95 percent hydrilla). Intermediate levels of preemptive establishment by *vallisneria* result in intermediate dominance.

Likewise, in pond research, *vallisneria* populations were able to effectively resist invasion by hydrilla and Eurasian watermilfoil only when allowed several months preemptive establishment.¹

The results of the field trials reported here are encouraging in that they demonstrate that the same principles apply in the field. As we learn more about how to efficiently establish native plants in the field and how to selectively control nuisance plants with herbicides or biological controls, we will come closer to our ultimate goal of providing our reservoirs with a healthy, resilient community of beneficial native plants.

American Lotus:Eurasian Watermilfoil Competition

American lotus is a native emergent/floating-leaved aquatic plant which has the potential to effectively compete with nuisance submersed plant species. Although an aggressive growing plant itself and not desirable in high-use areas, it may be useful for producing "gaps" within large monospecific beds of submersed exotic weeds like hydrilla and Eurasian watermilfoil.

Methods

Small plots of lotus were established within a large fenced enclosure at the Osa-Win-Tha site by plantings of scarified seed in 1991 (Doyle and Smart 1993a). By the summer of 1992 the lotus colony had filled the entire enclosure; it continued to expand from year to year in concentric circles. At the time of lotus establishment, Eurasian watermilfoil was not present in the cove, although the area has a history of milfoil infestation. In 1993 and 1994 there was regrowth of Eurasian watermilfoil in the area to such an extent that it required TVA control treatments. This regrowth pattern of milfoil in an area where a lotus colony was continuing to spread offered us the fortuitous opportunity to examine the ability of a developing lotus colony to withstand invasion by milfoil.

In August 1994 a harvest was made to quantify the regrowth of milfoil in relation to the degree of lotus establishment. Ten replicated 0.25-m quadrats were harvested in each of four zones within and near the lotus colony corresponding to varying degrees of lotus establishment (Figure 3). The interior zone was where the original plantings had been made and were therefore in the third/fourth year of lotus colonization. The next zone was the area to which the lotus colony had expanded to in 1993 (second year of colonization), while the third was the area to which the lotus had only recently expanded to (first year of colonization). The last zone was completely

¹ Unpublished research, R. M. Smart.

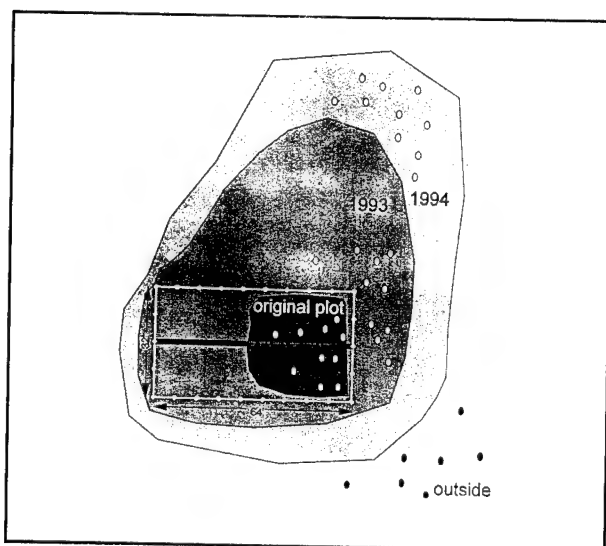


Figure 3. Approximate maximum summer distribution of the American lotus colony at Osa-Win-Tha cove. The plots were first established within the enclosure in 1991-1992. In 1993-1994 the colony was expanding rapidly outside the fenced area. Milfoil biomass harvests were made in August 1994 within each of the annual growth zones of the colony, which represent different degrees of lotus establishment

outside the lotus colony. The plant material collected was returned to the lab, sorted by species, and dried to constant weight at 60 °C.

Results and discussion

The harvest shows dramatic differences in milfoil biomass with respect to the degree of lotus establishment (Figure 4). In areas where lotus was well established (colony age 2 yr or greater), and presumably effecting competitive control over milfoil regrowth, the biomass of milfoil was very low. In sharp contrast, milfoil biomass was high outside the colony and in the area where lotus was just beginning to grow. These last two zones represent the regrowth of milfoil with little or no competitive interference from lotus.

These results reinforce the conclusions of Doyle and Smart (1994) that Eurasian watermilfoil is unable to expand beneath the canopy of an established lotus colony. Based

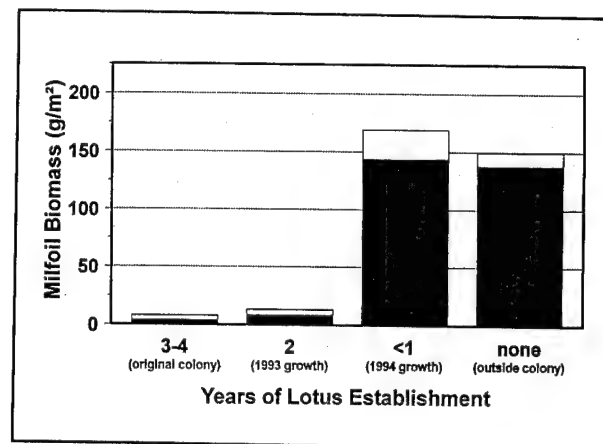


Figure 4. Eurasian watermilfoil mass in August 1994 within the various growth zones of the American lotus colony at Osa-Win-Tha cove

on observations of lotus and Eurasian watermilfoil growing together in experimental ponds at the LAERF, we anticipate that the lotus colony will continue to expand within the shallow water zone of the cove and displace Eurasian watermilfoil as it grows.

Pickerelweed:Lyngbya Competition

Background

Lyngbya is a nuisance, mat-forming cyanobacteria which degrades littoral habitats by the accumulation of high levels of mass. During the summers, much of this mass floats to the surface and forms a thick, foul-smelling, floating mat at the water surface within protected embayments. The result is that the lyngbya mat becomes stratified into three distinct layers: the floating surface mat, the benthic mat, and lyngbya buried down in the sediments. The heavy surface mat which develops during the summer, coupled with the summer water level fluctuations for mosquito control on Guntersville Reservoir result in a particularly harsh environment within which to establish emergent aquatic macrophytes. Previous studies (Doyle and Smart 1993a) demonstrated that pickerelweed was the best adapted for this environment.

Methods

Pickerelweed was established in 1992 in two small 1.3-m by 2.6-m plots within a larger enclosure at Waterfront site, an area completely dominated by thick mats of lyngbya. Transplants of dormant rhizomes collected from Reelfoot Reservoir, TN, grew well, flowered, and set seed during the first year of growth (Doyle and Smart 1993a). In the spring of 1993 many seedlings germinating within the enclosure, which, along with the planting of 50 additional transplants made in May 1993, resulted in an expansion of the pickerelweed plot to cover about 40 m² or 20 percent of the enclosure.

In August of 1993, thirty-two 0.25-m² quadrats were sampled within the fenced enclosure where the pickerelweed was established. Lyngbya biomass was collected from the 30 randomly selected areas of the enclosure and included samples from both the pickerelweed plots (10 samples) and unvegetated areas of the enclosure. Lyngbya biomass samples were collected separately from the various strata at which it was found (floating mat, benthic mat, and buried in the sediments). Lyngbya mass within vegetated and unvegetated sites was compared utilizing a t-test, after the sample variances were found to meet the homogeneity assumptions.

Results and discussion

After two growing seasons of pickerelweed at the Waterfront site, the mass of lyngbya within the vegetated plots was significantly lower than in the adjacent, unvegetated sites (Figure 5). Although the total mass of lyngbya within the vegetated plots was significantly lower than in unvegetated plots, most of this difference was due to the virtual lack of a floating mat within the vegetated area.

The mechanism for the reduction of lyngbya mass is unclear in the present study. However, it is unlikely to have simply died and decomposed. Due to the extremely thick

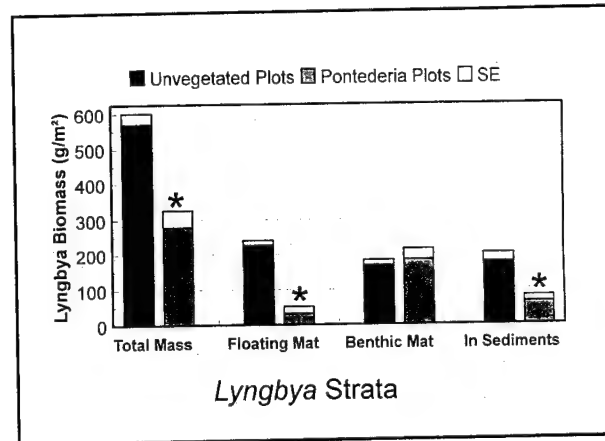


Figure 5. Average (\pm SE) lyngbya mass found at the various strata at which it was found vegetated (dark bars) and pickerelweed plots (cross-hatched bars). Significant ($p < 0.05$) differences between unvegetated and pickerelweed plots are indicated with an asterisk

sheaths of lyngbya, the decomposition half-life of freeze-dried cells is approximately 20 months.¹ Even so, in the long run, macrophytes may be able to change the littoral environment so that it is much less suitable for lyngbya growth by shading and reducing the nutrients diffusing out of the sediment to the lyngbya mat. Light measurements beneath the pickerelweed canopy showed that the emergent leaves reduced light availability by about 95 percent. In addition, the root uptake of the macrophytes reduced the nutrient levels in the sediments beneath the lyngbya mat during the growing season.

These data point to the potential for planting emergent macrophytes to ameliorating the negative impacts of the nuisance, floating mats of lyngbya which develop during the summer. Although this technique is unlikely to quickly eradicate the lyngbya, it should provide immediate relief from the nuisance floating mats which are common during the summer. In addition, by shading the mat and reducing the nutrients available from the sediments, the macrophytes may ultimately reduce the mass of lyngbya.

¹ Personal communication, B. Speziale, Clemson University, Clemson, SC.

Conclusions and Recommendations

As ecological "weed species" (that is, ruderal or r-selected), Eurasian watermilfoil and hydrilla are well adapted to quickly colonize disturbed or unvegetated environments. However, they may be less competitive than generally thought against well-established native plants. The data from these very important field trials provided additional evidence for the potential of utilizing native species to protect our reservoir systems from invasions of these nuisance exotics. Future research should focus on:

- Identifying additional competitive native species
- Developing efficient propagation and establishment techniques for native plants
- Conducting additional field trials utilizing longer time and larger space scales.

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Fish and Aquatic Plant Technology

Fish and Aquatic Plants: Overview of a New Technology Area

by
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Introduction

A new technology area in the APCRP began in FY95 titled Fish and Aquatic Plants. The overall objective of the technology area is to develop quantitative relationships between aquatic plants and fish community structure for broad applicability within geographic regions. These relationships will be used to help the Corps of Engineers identify plant management strategies that benefit the fishery. The technology area considers descriptive and functional aspects of plants by addressing three questions:

- (1) What types of fish and life stages are associated with aquatic plants?
- (2) How do plants influence feeding, growth, and reproduction of fish?
- (3) Are there optimum densities and spatial configurations of plant beds that provide greatest fishery benefits?

Description of Work Units

Two work units in the technology area are currently funded: (1) Habitat Classification of Aquatic Plants for Fish and (2) Reproduction of Fish in Aquatic Plant Habitats. Three additional work units have been, or will be, proposed: (1) Food Web Interactions, (2) Growth and Condition of Fishes, and (3) Strategies for Fisheries Management in Vegetated Water Bodies. The last work unit on management strategies will compile information obtained from other work units, demonstrate fish/plant relationships emphasizing limiting factors, and produce workable models that plant managers can use to simulate and predict fishery benefits of plant control operations (Killgore,

Dibble, and Hoover 1993). An example of this type of research is described by Storlie et al. (1995).

The Habitat Classification work unit is evaluating a standardized system of classifying habitats created by aquatic plants and documenting fish distribution, diversity, and abundance in each habitat. A hierarchical approach to classify plant habitats is being applied (Dibble and Killgore 1994): regional, system, and local.

Fish-plant interactions will vary by geographical region, thus a regional approach is required to identify different solutions to a particular problem. Three broad regions of the United States that have either different plant growth patterns, different fish species assemblages, or a combination of both are being considered (Dibble and Killgore 1994): Southeastern, Northeastern, and Pacific Northwest.

A system-level classification considers within-water-body characteristics of the plant bed. To evaluate this aspect, multispectral videography has been obtained of aquatic plants in Caddo Lake, TX, and is being used to map spatial configuration of plant beds (Hardy 1995). Synoptic studies of fish composition within delineated plant habitats will be conducted during FY95. Furthermore, the effect of macrophytes and fish on coupling of the limnetic and benthic zones is being considered as an important factor in these system-level studies (Drenner and Gallo 1995).

Classification of plants on a local scale considers underwater structural heterogeneity. Once individual patches of plants are identified and classified with aerial photography, spatial complexity is measured and related to

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

some biological function such as fish foraging (Dibble and Harrell 1995). These types of fish behavior studies are critical to understanding how fishes respond to different levels of structural complexity and, ultimately, to plant control.

Future Studies

Three studies are planned for FY95:

- (1) Reproduction of fish associated with aquatic plants will be evaluated in Caddo Lake during spring and summer 1995. Based on habitat classification from multispectral videography, larval fishes will be sampled in discrete plant habitats during the reproductive season. Composition and abundance of larval fishes will be compared across habitats to evaluate preference in rearing habitat. These data will be compared to a nonvegetated lake (Hugo Lake, OK) where similar studies are being conducted.
- (2) A pond experiment of factorial design will be conducted to examine how fish and macrophytes interact to couple the limnetic and benthic zones of lakes. As part of this study, effects of turbidity and macrophytes on efficiency of electrofishing and popnets used to collect fish in vegetated areas will be measured.
- (3) A comparative study of fish-plant interactions between oligotrophic and

eutrophic lakes in Wisconsin will be developed. All lakes are colonized by Eurasian watermilfoil, but fertility differs substantially. System- and local-level studies will begin and continue for several years.

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Use of Multispectral Videography with GIS for Spatial Relationships Between Aquatic Vegetation and Fish

by
Thomas B. Hardy¹

Introduction

Quantifying of relationships between aquatic plant habitats and fish production represents an important element in the on-going APCRP of the Corps of Engineers at the WES. The research is intended to explore these relationships at the microscale which can then be generalized to allow extrapolation and inferences at the mesoscale within lake and reservoir systems. The results will be used in the decision-making process for aquatic plant control strategies.

This paper describes a collaborative effort between WES and Utah State University on the application of remotely sensed data using digital multispectral videography and GIS to support field sampling efforts, data tracking and analysis, and extrapolation of microscale results over larger mesoscale study areas at Caddo Lake, TX. The specific objectives are to evaluate the use of multispectral videography for the delineation of aquatic and emergent vegetation classes within Caddo Lake, provide a spatial linkage between ground based field measurements of larval fish use by specific vegetation classes, and to use GIS to integrate fish and vegetation relationships developed at the microscale to spatially extrapolate fish and vegetation class dependent relationships at the mesoscale for large areas within Caddo Lake.

Multispectral Videography Image Acquisition System and Image Processing

Multispectral videography system

The aerial multispectral videography system used for data collection was developed at

Utah State University (Neale 1992). This system consists of three COHU 4810 series high resolution (525 horizontal lines) monochrome CCD video cameras with 10 nm interference filters centered on 550nm (green), 650nm (red), and 850nm (infrared) regions of the electromagnetic spectrum. In this application, the system used 16mm SONY television quality lens and three PANASONIC AG-7400 S-VHS video tape recorders (425 horizontal lines) for recording camera output during image acquisition. In addition, the instrument package also contained an Exotech 4-band radiometer with Landsat Thematic Mapper bands TM1 (450-520 nm), TM2 (520-620 nm), TM3 (630-690 nm), and TM4 (760-900 nm) with square 1-degree field-of-view lens, an Everest thermal infrared radiometer (8-14 m) with a circular 2-degree field-of-view, and an OMNIDATA 700 series Polycorder datalogger for recording DC camera voltages and analog signals from the two radiometers. Image frame control and geo-location were accomplished with the inclusion of a TRIMBLE Pathfinder global positioning system (GPS) with output recorded by an OMNIDATA 600 series datalogger and a SMPTE time code generator to provide time and frame counts for later image digitization.

The cameras and radiometers were mounted in a vibration isolation frame to minimize blurring. Camera lens focuses were set at infinity and the focal axes aligned to converge on an object at the expected height above ground for data collection. All three cameras were synchronized with the green camera designated as the master thus allowing all three cameras to simultaneously record the same image. The synchronization signal from the

¹ Director, Institute for Natural Systems Engineering, Utah Water Research Laboratory, Utah State University, Logan, UT.

master camera was fed to the time code generator to supply time/frame code information which was recorded on audio channel #2 by each video recorder. The time/frame code information was also provided in a data block on the bottom of the green camera image for visual reference during image digitizing. Another data block on the bottom of the red camera image contained GPS information from the TRIMBLE Pathfinder.

Image preprocessing

Images were viewed and digitized with a PANASONIC S-VHS AG-7500 editing machine controlled via a DIAQUEST board and software system on a PC-386 (Neale 1992). The DIAQUEST board used the time code to locate the same scene on each monochrome video tape for automatic digitization of desired frame sequences. Individual frames were digitized to an 8-bit (256 gray scales) file using a TARGA+ board for transfer and processing on an IBM RS-6000 530H workstation. Data processing on the workstation was conducted using the Earth Resource Data Analysis System (ERDAS) and software developed for this project.

Images were first corrected for vignetting via procedures established by Crowther (1992). Vignetting is the darkening of the image with increasing distance from the center of the lens. The vignetting correction adjusts the imagery for departures from perfect focusing which causes this effect as well as accounting for nonuniformities in the camera sensing chip. Three-band false color composite images of each video scene were constructed by registration of the red and infrared images to the green image. Figure 1 provides an example of each single monochrome band and the final 3-band false color composite for Caddo Lake. Full-scale photographic quality prints of single image scenes were subsequently printed for use in field reconnaissance.

The interlaced scanning resulting from the RS-170 television standard resulted in a horizontal shifting due to aircraft motion perpen-

dicular to the flight path (transitional and roll effects) and a vertical shifting due to both forward plane motion and pitch. Image shifts due to platform yaw were not corrected. Vertical line shifting was corrected by maximizing the correlation of the summed brightness values for shifting of odd/even lines (Neale et al. 1993). Every tenth value in an image centered window 1/9th the size of the image was used in the correlation. Horizontal line shifting was then performed in the same manner. Images were then visually reviewed to confirm the selected shifting.

Image classification

The individual 3-band images were clustered and classified using statistical clustering with a Bayesian/maximum likelihood classification (ERDAS 1991). Image classification was performed with the ERDAS software package on an IBM RS/6000 Model 530H. The resulting GIS image files were stitched to form mosaics for selected target large-scale study sites within Caddo Lake based on prior field reconnaissance.

Statistical clustering

This algorithm steps nonoverlapping 3 by 3 windows across the image and performs a spatial homogeneity test to determine a user-specified number of signatures, N . The standard deviation, s_i , of each band for each window is compared against a lower bound. This lower bound prevents ill-conditioned covariance matrices in the classification routines which are used with the signatures generated (covariance matrices for the classification algorithms are calculated as described above for the transformed divergence). If this test is passed, s_i is tested against an upper bound, G , to determine its use in fixing the clusters (Equation 1).

$$G = \text{maximum } (U, m_i \cdot V/100) \quad (1)$$

where U is a user-defined value to ensure windows with low means are not discounted as homogeneous, m_i is the mean for band i in the

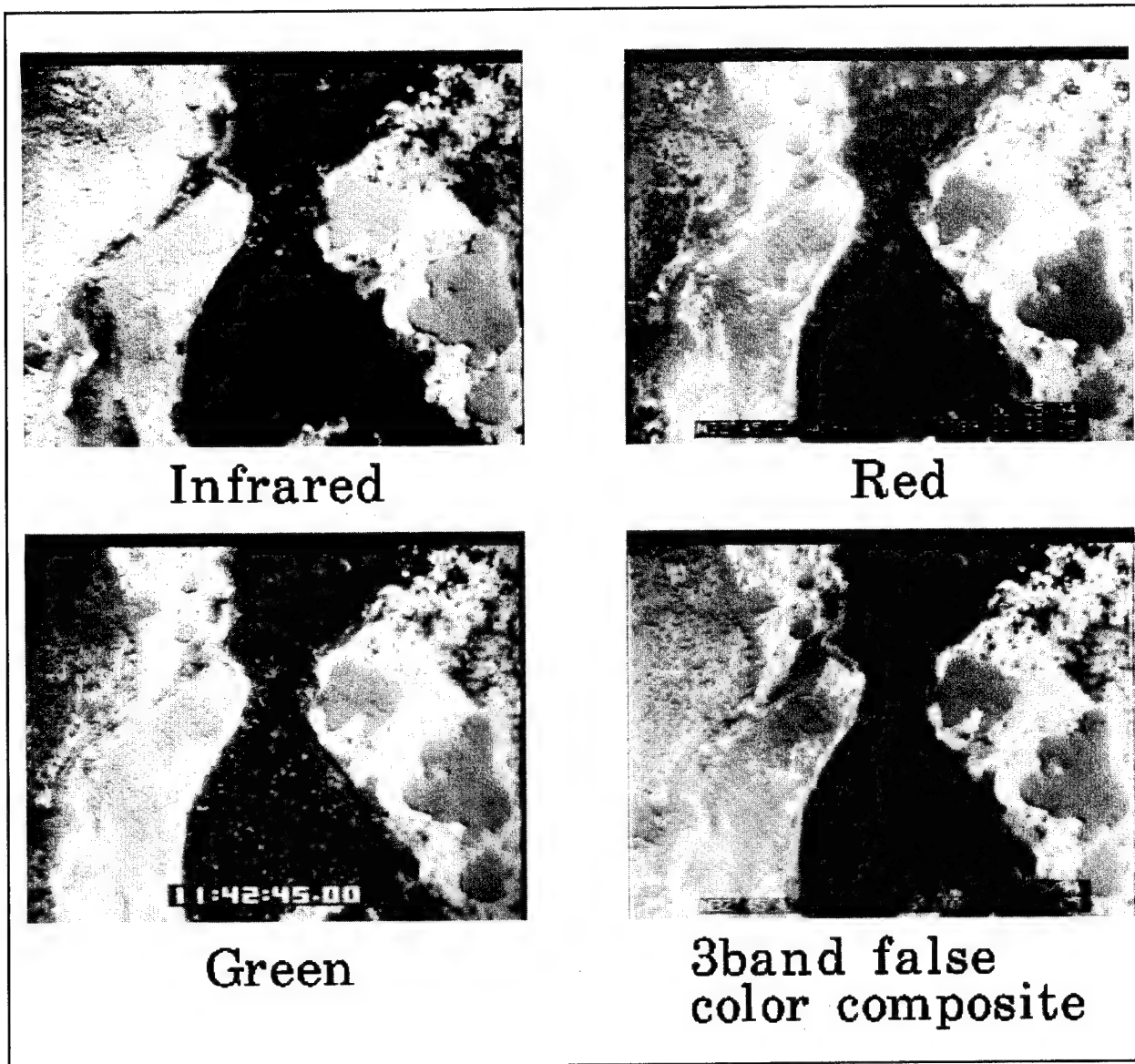


Figure 1. Example of individual monochrome digital images filtered in the infrared, red, and green ranges of the electromagnetic spectrum and rectified 3-band false color composite (printed as monochrome for publication)

window, and V is the user-selectable coefficient of variation (percentage) used with the mean as an alternative upper limit. If $s_i < G$, the window is considered homogeneous in band i , and if all bands are homogeneous, the window is used in cluster determination. Each window is initially considered a separate cluster until $N + 1$ clusters are found. Windows are pair-wise compared and those whose scaled spectral distance, S_x , (Equation 2) exceeds a user-specified maximum are merged (ERDAS 1991).

$$S_x = \sqrt{\sum \frac{(\mu_{ai} - \mu_{bi})(W_{ai} + W_{bi})^2}{0.05(W_{ai}\mu_{ai} + W_{bi}\mu_{bi})}} \quad (2)$$

where

W_{ai}, W_{bi} = number of windows in cluster a, b band i

μ_{ai}, μ_{bi} = mean of cluster a, b band i

Based on prior field site visits, the user-defined upper limit for number of classes was

set to 20. The resulting clusters were submitted to transformed divergence testing and yielded 10 to 20 classes for each 3-band composite image within the selected large-scale study areas. The resulting training sets were then used for image classification with a Bayesian/maximum likelihood classification decision rule (Duda and Hart 1973; ERDAS 1991).

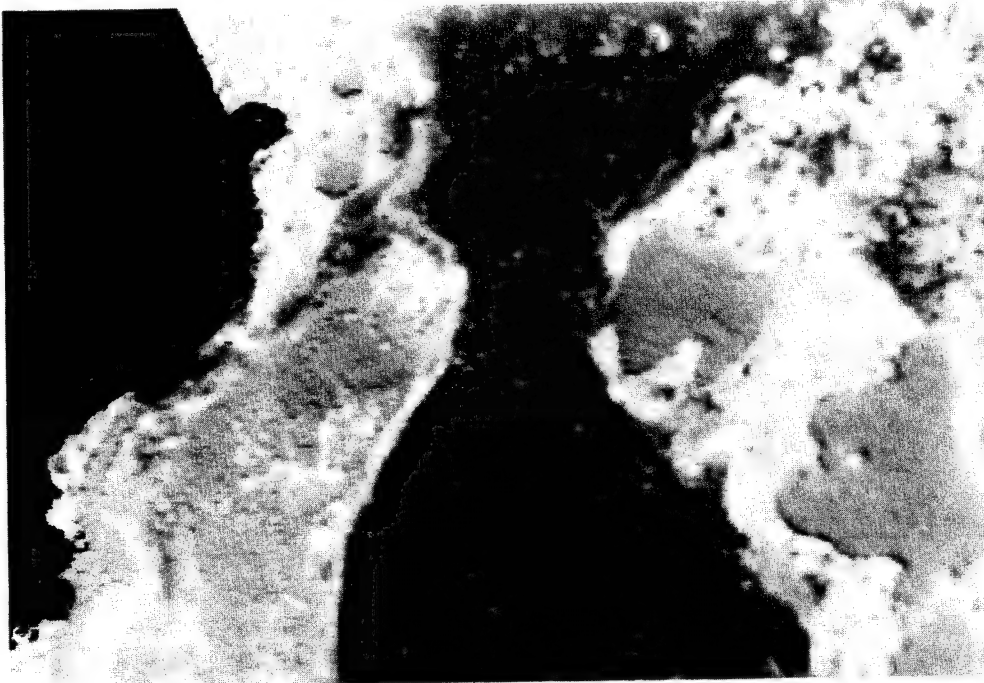
Field Reconnaissance and Ground-Based Validation of Vegetation Classifications

An example of a single 3-band false color composite and statistical clustering results for classified vegetation is provided in Figure 2. The "black" area along the left-hand side of the image has been masked out to highlight features within the remaining image. Photographic quality color prints of the 3-band composite images and the clustering results for vegetation classes were utilized in the field to assist in the association of computer-generated vegetation classes with actual vegetation types within the field. These images were also utilized as navigation aids and spatial orientation while conducting field investigations. For example, note the dark linear feature along the right-hand margin of the "hour glass" in Figure 2, which is a boat lane cut through the vegetation within this area of Caddo Lake. Dense beds of submerged aquatic vegetation are indicated by lighter grey shades within the "hour glass," while individual cypress trees are easily distinguished as "dots" in the images. Dense stands of emergent and floating vegetation mats surround the two "islands" along the right-hand margin of the boat lane in this image.

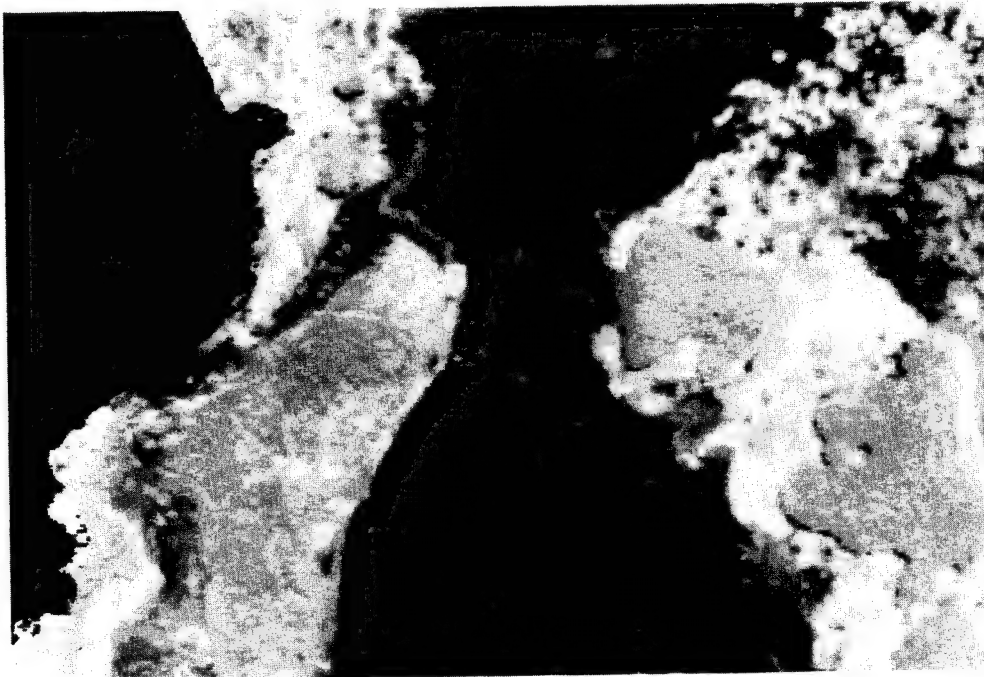
Spatial resolution of the digital imagery was 1.5 meters/pixel and at this resolution, location of specific trees and other aquatic vegetation features could be determined to within approximately 2 meters during field reconnaissance of the study sites within Caddo Lake. Utilization of the imagery during repetitive site visits is anticipated to help reduce spatial sampling errors associated with delineation of specific sampling sites throughout the study area.

The individual 3-band composites covering the spatial extent of each target large-scale study area were also stitched into a single mosaic for use in field investigations and subsequent image classification for vegetation categories. Figure 3 represents an example of a single mosaic using over 20 individual overlapping single 3-band composite images for the Carter Lake study area from Caddo Lake. The area represented by Figure 2 is located in the upper left quadrant of Figure 3. These larger scale mosaic images were also useful in orientation during field work and for examination of the spatial relationship of different types and densities of vegetation features in relationship to broader spatial features within the study area.

In general, the spectral characteristics for emergent and floating vegetation types versus submerged aquatic vegetation classes were found to be consistent for imagery throughout the target large-scale study areas. Spectral signatures associated with vegetation classes derived from unsupervised statistical clustering procedures resulted in consistent delineations between cypress, duck weed, water lily, and submerged pond weed/milfoil. Successful computer classification is a result of the inherent differences in spectral properties and spatial heterogeneity of both emergent and submerged aquatic plant beds illustrated in Figures 2 and 3, although these differences were not always apparent during ground-based field observations. Ground truth data are presently being utilized to undertake supervised classification of the imagery to improve vegetation class separation over each of the large-scale study areas. Revised classification results will then be field validated in areas where initial ground truth data are not collected. Image classification errors will then be assessed for each of the vegetation classes to assist in the interpretation of extrapolated results. Research at Utah State in coastal wetland sites with spatial and spectral properties similar to this imagery has shown that development of spatial and spectral metrics can successfully be utilized in the characterization of plant community classification and delineation of spatial heterogeneity (Neale, Hardy, and Shoemaker 1994).



3band composite



**Example classified emergent
vegetation and open water**

Figure 2. Example of 3-band false color composite and corresponding statistical clustering of vegetation classes based on spectral properties (printed as monochrome for publication)



Figure 3. Example of large-scale study area mosaic of 3-band composite video imagery for the Carter Lake area of Caddo Lake (printed as monochrome for publication)

Integration of Imagery and Field Sampling Within GIS

The digital 3-band false color imagery and classified image results have been imported to the ArcInfo GIS system at Utah State Uni-

versity. The GIS system is presently being utilized to register this imagery to USGS Quad maps of the study area to permit geo-referencing of both the image features as well as location of specific field-based sampling points. At present, it is anticipated the photographic

quality printed imagery of the 3-band composites and classified imagery will be utilized during larval fish sampling to identify the actual spatial location of each sample on the images during field work. The sample locations will then be included directly within the GIS by simply locating the same area within the imagery on the GIS.

Supplemental multispectral video imagery will be collected this winter (1994-1995) at a spatial resolution of approximately 40 to 50 cm per pixel. This fine-scale imagery will significantly reduce the spatial location errors and is anticipated to improve the vegetation classification results since these images can be combined with the initial multispectral imagery to take advantage of temporal characteristics of spectral signatures for many of the target vegetation species. The new imagery will also be registered to both the 1.5-meter resolution images and the USGS Quad map base for use in the larval fish study.

Each sample location will be linked within the GIS which associates delineated vegetation classes to a database containing the physical, chemical, and biological attributes collected at each sampling point. These linked data will be used in mesoscale extrapolations of larval fish distribution and abundance over the larger scale study; additional field sampling will validate the extrapolations. If this effort is found to produce adequate extrapolations based on field validation tests, then the GIS can be used to assess the potential impacts and management implications of various aquatic plant control strategies by simulating spatial changes in vegetation attributes and comparing pre- and post-control strategies.

Conclusions

Use of multispectral video imagery has been shown to provide an excellent tool to evaluate spatial characteristics of large-scale study areas within Caddo Lake. Sample locations for larval fish collections can be quantitatively evaluated with respect to spatial heterogeneity of aquatic plant beds. Statistical clustering of the digital imagery was shown to provide consistent classification of key vegetation types

which included both emergent and submerged vegetation communities. Imagery was also found to provide good delineations of the spatial heterogeneity of emergent and submerged aquatic vegetation beds based on differential spectral properties within the imagery. The imagery facilitated spatial orientations within the field to within 2 meters, and provides an excellent tool for accurate linkage of field-based measurements of physical, chemical, and biological data to specific localities. The use of the digital imagery in conjunction with field-based sampling efforts within GIS will be evaluated as a means to extrapolate relationships obtained at the microscale to assessments at the mesoscale for larger scale study areas within Caddo Lake. Field validation studies of the extrapolations are planned as part of the study design.

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Experimental Pond Study of the Effects of Carp and Fertilizer on Water Quality, Macrophytes, and Centrarchids

by

Kirsten L. Gallo¹ and Ray W. Drenner¹

Introduction

Studies have shown that fish can have significant effects on the water quality of lakes (Northcote 1988; Drenner, Smith, and Threlkeld, in press) with most investigations focusing on the impacts of zooplanktivorous fish. Ecologists have hypothesized that indirect enhancement of phytoplankton biomass by zooplanktivorous fish diminishes with increased nutrient loading (McQueen, Post, and Mills 1986; but see Lancaster and Drenner 1990; DeMelo, France, and McQueen 1992). Specifically they hypothesized that an interrelationship exists between fish and nutrients (nitrogen (N) and phosphorus (P)) so that the effects of fish on phytoplankton are significant in oligotrophic systems but not in eutrophic systems. This hypothesis implied that lake management strategies aimed at reducing eutrophication could ignore the ecological effects of fish.

However, the biomass of fish communities of reservoirs in the central and southern United States are dominated by omnivorous fish such as gizzard shad (*Dorosoma cepedianum*) and carp (*Cyprinus carpio*) and not obligate zooplanktivorous fish (Miranda 1983). We define omnivorous fish as those that feed on detritus, phytoplankton, periphyton, and invertebrates. Omnivorous fish not only enhance phytoplankton biomass through suppression of herbivorous zooplankton (Drenner, Threlkeld, and McCracken 1986) but also through transfer of nutrients from the benthic zone to the limnetic zone (Lamarra 1975; Brabrand, Faafeng, and Nilssen 1990). Omnivorous fish act as "nutrient pumps"

when they consume sediment-bound nutrients and subsequently excrete them into the water column. Also clay particles with adsorbed nutrients may be resuspended by the fish's feeding activities. Nutrients transported into the water column through excretion and sediment resuspension are available to planktonic communities. Unlike zooplanktivorous fish whose suppression of macrozooplankton often does not cascade to phytoplankton (McQueen 1990; DeMelo, France, and McQueen 1992), studies of the impacts of omnivorous fish almost always show that omnivorous fish enhance phytoplankton biomass and/or primary productivity, even in eutrophic systems (Drenner, Smith, and Threlkeld, in press).

Two experiments have examined how the effects of omnivorous fish change with nutrient loading. Riemann (1985) conducted an enclosure experiment of factorial design in which two levels of omnivorous fish (presence and absence of roach (*Rutilus rutilus*)) were cross-classified with two levels of nutrient loading (presence and absence of nutrients (N+P)). Riemann (1985) found that enhancement of chlorophyll by roach was greater in the treatment combination with highest nutrient loading. Drenner, Smith, and Threlkeld (in press) conducted a tank-mesocosms experiment of factorial design in which five biomass levels of omnivorous gizzard shad were cross-classified with two levels of lake trophic state achieved by filling tank-mesocosms with water and plankton transported by truck from an oligotrophic lake and a eutrophic lake. The enhancement of chlorophyll *a* by gizzard shad was dependent on lake trophic state and was most intense in the mesocosms with

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water from the eutrophic lake. Based on this experiment, Drenner, Smith, and Threlkeld (in press) hypothesized that omnivorous fish may interact synergistically with lake trophic state so that the limnological effects of omnivorous fish become more intense with increased eutrophication.

Here we present the results of a 23-pond experiment testing the hypothesis that the effects of omnivorous fish intensify with nutrient loading. The experiment consisted of a factorial design with two levels of omnivorous carp cross-classified with two levels of fertilization. This experiment allowed us to evaluate the separate and combined effects of omnivorous fish and fertilizer on water quality, macrophytes, and centrarchids.

Methods

The experiment was set up in fall 1991 and winter 1992 at the Eagle Mountain Fish Hatchery, a facility leased by Texas Christian University from the Tarrant County Water Control and Improvement District Number One. The factorial design (presence and absence of carp cross-classified with presence and absence of fertilizer) consisted of four treatment combinations (carp absent and no fertilization, carp present and no fertilization, carp absent and fertilization, carp present and fertilization). Each treatment combination had five or six replicate ponds.

In the experiment we used carp as a representative of a species in the guild of omnivorous fish present in U.S. reservoirs. Carp feed on a mixture of macrozooplankton, detritus, and benthic invertebrates (Gurzeda 1960; McCrimmon 1968; Ulaiwi 1990). Carp have been found to affect various components of aquatic communities including zooplankton, phytoplankton, benthic invertebrates, and macrophytes (Cahn 1929; Threinen and Helm 1954; Hruska 1961; Grygierek 1962; Losos and Hetesa 1973; Lellak 1978; Crivelli 1983; Richardson, Wickham, and Threlkeld 1990).

Ponds were stocked with fish from October 1991 through February 1992 with 11 of the 23 ponds receiving approximately 1,200 fingerling carp/ha. The carp ponds were also stocked with gizzard shad, but apparently the dense vegetation may have lowered gizzard shad survival and they became established in only one pond.

Because centrarchids (bluegill (*Lepomis macrochirus*) and largemouth bass (*Micropterus salmoides*)) always occur in lakes containing omnivorous fish, all ponds were stocked with fingerling bluegill (1,200/ha) and largemouth bass (120/ha) during fall 1991 according to guidelines of the Texas Chapter of the American Fisheries Society (1986). In this experiment we treated bluegill and largemouth bass as response variables and did not test for the effects of bluegill and largemouth bass whose ecological impacts have been previously studied (Hambright et al. 1986; Lazzaro et al. 1992).

Prior to the experiment, we observed that many of the ponds contained dense populations of macrophytes. In an attempt to reduce macrophyte populations, we stocked sterile triploid grass carp (*Ctenopharyngodon idella*) (25/ha) into all ponds on 21 November 1991. However, macrophytes remained abundant in most ponds as found by Kirk (1992).

Fertilization consisted of adding both phosphorus and nitrogen, the two nutrients often limiting primary productivity and fish production of lake systems (Elser, Marzolf, and Goldman 1990; Boyd 1981). Previous studies by the Texas Parks and Wildlife Department using the ponds at Eagle Mountain Hatchery showed that fertilization with phosphorus and nitrogen increased plankton production.¹ Based on discussions with Farquhar, we applied liquid fertilizer (10N:34P:OK) (15.8 to 19.9 kg/L) at a rate of 9.4 L fert./ha. In 1992 fertilizer was added every 2 weeks to the ponds from 20 March to 29 March. In 1993 we added fertilizer every 2 weeks from 18 March to 25 June and monthly from July through September.

¹ Personal communication, 1994, B. Farquhar, Biologist, Texas Parks and Wildlife Department, San Angelo, TX.

Ponds were sampled on 20 August 1993 to assess turbidity, chlorophyll *a*, total phosphorus, and total nitrogen. Integrated water-column samples for water quality analyses were taken with a PVC tube. Turbidity was measured with a model 2100A Hach turbidimeter. To determine chlorophyll *a*, we filtered water through 0.45- μ m Millipore filters, wrapped the filters in aluminum foil and froze them. Chlorophyll *a* was extracted in 2:1 chloroform-methanol solution in the dark for a minimum of 4 hr and read at 665 nm (Wood 1985). Samples for total phosphorus were digested with potassium persulfate (Menzel and Corwin 1965) and analyzed by a modification of the malachite green method (Van Veldhoven and Mannaerts 1987) in which 1 ml of color reagent was added to 5-ml subsamples and read at 610 nm. Samples for total nitrogen were digested with alkaline potassium persulfate (D'Elia, Steudler, and Corwin 1977) and analyzed by UV estimation at 220 nm (APHA 1985).

Between 30 July and 16 August 1993 ponds were sampled to estimate the biomass of macrophyte species. Twelve samples were taken per pond along a line-transect running from near the water inflow area to the opposite side of the pond. One sample was taken 0.5 m offshore and the other 11 samples were taken at randomly selected distances along the transect. At each site all vegetation inside a 0.023- m^2 area (15 cm by 15 cm) was removed by hand. Macrophytes were placed in nylon-mesh bags and stored in water until they were sorted to species in the lab. Specimens were blotted with paper towels, dried in the sun for a few days, and weighed to the nearest 0.01 g.

Electrofishing was conducted 29 October and 13 November 1993 during the daylight hours to assess the fish populations in the ponds. The electrofishing unit consisted of a 3,750-W generator and a pulsator which produced a 10-amp current. Electrofishing was conducted along the perimeter of each pond for approximately 10 minutes. One person on the bow of the boat dipped fish from the surface of the water. The fish were placed in stock tanks until their weights and total

lengths were taken, then they were returned to the ponds.

The data set was analyzed by ANOVA for cross-classified factors (Winer 1971). The analysis allowed us to examine main effects and interaction effects. Main effects are the impact of each treatment factor (carp and fertilization) averaged across all other treatment conditions, while interaction effects are the amount of measured variation in the response variables that is due to interdependence among treatment factors. It is the interaction effect term that allowed us to assess if the effects of the carp were independent of fertilization. All analyses of variance were completed with MYSTAT. To avoid making a Type II error (failing to reject a false null hypothesis), we used $\alpha = 0.10$ to determine statistical significance.

Results

Chlorophyll *a* and turbidity were increased in the presence of carp (Figure 1). The addition of fertilizer significantly increased chlorophyll *a* and total phosphorus, but had no effect on other response variables. We detected a significant carp \times fertilizer interaction effect for chlorophyll *a*. Carp increased chlorophyll *a* levels more in the presence of fertilizer indicating that the effects of carp are enhanced under eutrophic conditions.

Eleven species of macrophytes were found in the ponds (Figure 2). The most abundant species included Eurasian watermilfoil (*Myriophyllum spicatum*), American pondweed (*Potamogeton nodosus*), American lotus (*Nelumbo lutea*), and bushy pondweed (*Najas guadalupensis*). Those species occurring in fewer than five ponds include *Chara* sp., narrowleaf pondweed (*Potamogeton obtusifolius*), smartweed (*Polygonum amphibium*), and coontail (*Ceratophyllum demersum*). The presence of carp reduced the biomass of *N. guadalupensis*, filamentous algae, and the number of macrophyte species per pond (Figure 3). No significant fertilizer effects or carp \times fertilizer interaction effects were detected on the macrophytes.

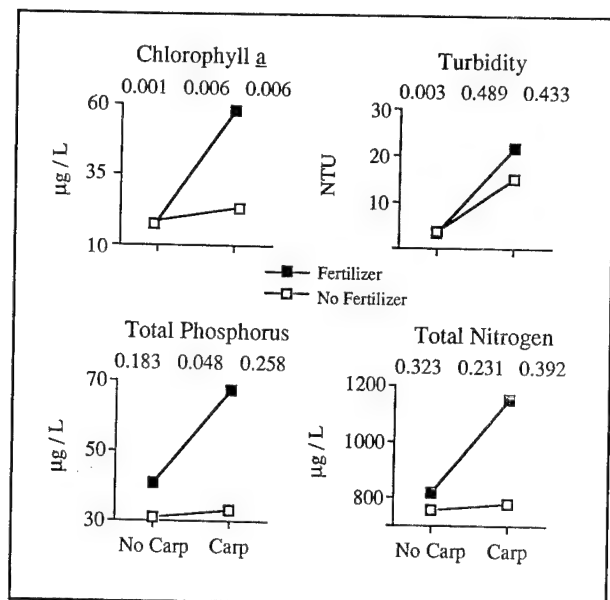


Figure 1. Mean responses of water quality variables to carp and fertilizer. ANOVA probability values are given for the main effects of carp (left), fertilizer (center), and carp \times fertilizer interaction effect (right)

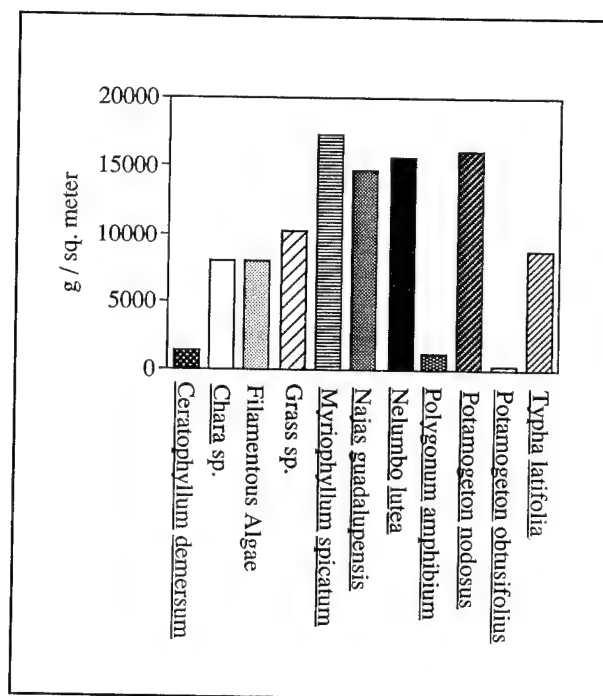


Figure 2. Total biomass of macrophytes in ponds at Eagle Mountain Fish Hatchery

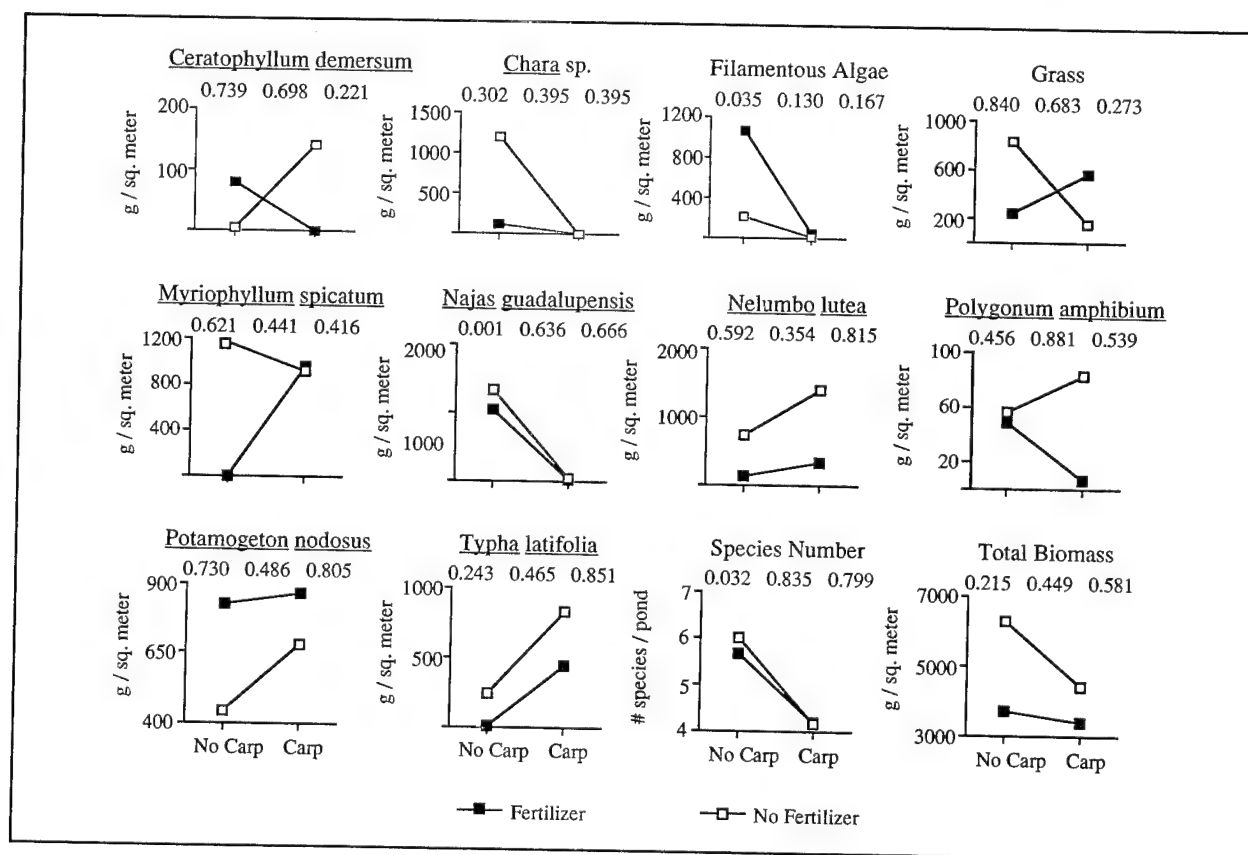


Figure 3. Mean responses of macrophytes to carp and fertilizer. ANOVA probability values are given for the main effects of carp (left), fertilizer (center), and carp \times fertilizer interaction effect (right)

The macrophytes may have "dampened" the effects of carp on the water column. Carp-mediated increases in turbidity, total phosphorus, and chlorophyll *a* appeared to diminish with increased biomass of Eurasian watermilfoil (Figure 4) suggesting that milfoil may regulate the effects of carp on the water column.

The presence of carp significantly reduced the biomass of bluegill and largemouth bass collected by electrofishing (Figure 5). No fertilizer effects or carp \times fertilizer interaction effects were detected for bluegill or largemouth bass. We cannot evaluate whether the lower catch rates of bluegill and largemouth bass were caused directly by carp or reflect a

decrease in the capture efficiency of electrofishing due to increased turbidity in the carp ponds.

Discussion

In this pond experiment the presence of carp increased chlorophyll *a* with the most dramatic effects occurring in the fertilized ponds. These results are consistent with the hypothesis of Drenner, Smith, and Threlkeld (in press) that the effects of omnivorous fish act synergistically with nutrient loading to control phytoplankton biomass. The implication of this fish-nutrient interaction is that lake managers combating eutrophication must

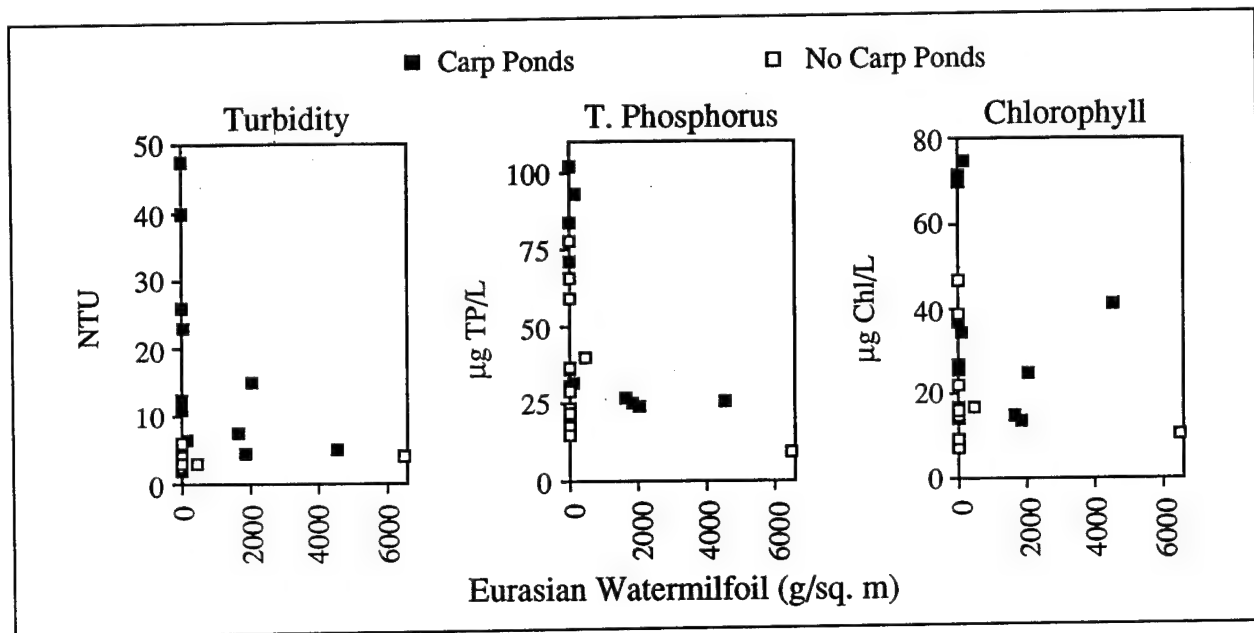


Figure 4. Changes in turbidity, total phosphorus, and chlorophyll *a* in the ponds as a function of biomass of Eurasian watermilfoil. Points represent individual ponds sampled in summer 1993

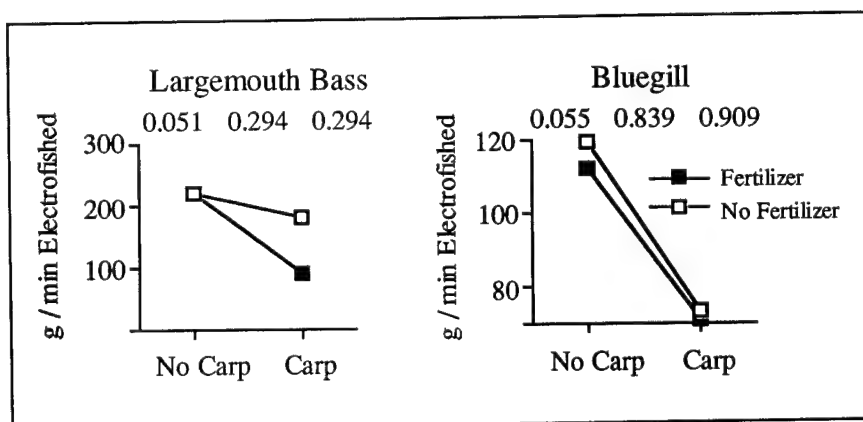


Figure 5. Mean responses of bluegill and largemouth bass to carp and fertilizer. ANOVA probability values are given for the main effects of carp (left), fertilizer (center), and carp \times fertilizer interaction effect (right)

take into account the presence of omnivorous fish (Lee and Jones 1992).

Although studies have found complex ecological relationships between fish, water clarity, and macrophyte populations (Killgore, Morgan, and Hurley 1987; Balls, Moss, and Irvine 1989; Irvine, Moss, and Balls 1989; Van Donk, Gulati, and Grimm 1989; Sondergaard et al. 1990; Hanson and Butler 1994), no studies have addressed how macrophytes may mediate the effects of omnivorous fish on lake water quality. Our pond experiment suggested that Eurasian watermilfoil may dampen the effects of bottom-feeding carp. In future studies we hope to examine how omnivorous bottom-feeding fish and macrophytes interact to couple the limnetic and benthic zones of lakes and regulate water quality and fish production.

Acknowledgments

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Some Macrophyte, Invertebrate, and Fish Associations in a *Myriophyllum spicatum* Lake: Will Increasing the "Edge" Change Anything?

by

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Introduction

Since the introduction of the exotic macrophyte Eurasian watermilfoil, *Myriophyllum spicatum*, dense macrophyte cover has become a problem in many Wisconsin lakes, including Fish Lake. In addition to limiting lake access, dense Eurasian watermilfoil beds have deleterious effects on fish behavioral and population characteristics. The stability and sustainability of fisheries depend on strong linkages to macrophytes, which provide habitat for early life stages of fishes, refuge from predators, and food resources (invertebrates). Macrophyte biomass and species composition are affected directly by management techniques such as harvesting and herbicides. Littoral zones are known to be important for fisheries, but lack of quantitative information has hampered efforts to integrate macrophyte management into fisheries management plans (Engel 1985).

Submersed macrophytes play a major role in aquatic ecology (Carpenter and Lodge 1986; Pardue and Webb 1985; Sculthorpe 1985). Aquatic plants serve as refuge for small fishes and provide important structural habitat for periphyton (Cattaneo 1983; Cattaneo and Kalff 1980; Morin 1986) and associated macroinvertebrates (Menzie 1980; Pinder 1986; Rosine 1955; Soszka 1975a, b). The macroinvertebrates are used as food by many fish species (Fairchild 1982; Keast 1984; Crowder and Cooper 1982; Pardue and Webb 1985). Density and morphology of vegetation can mediate predator success on invertebrates (Dionne and Folt 1991) and fish (Savino and Stein 1989, De Nie 1987). The

importance of macrophyte beds to specific fish assemblages is intuitively understood; the mechanisms and limitations of these relationships, however, are not so clear.

The Fish Lake project is interested in quantifying the effects of mechanical harvesting of dense macrophyte beds on fish population characteristics, such as growth and fitness. The manipulation of the dense milfoil beds is unique in that the macrophytes are harvested from the sediment surface to the water surface, thus creating deep channels from shore to the pelagia, and increasing the edge area of the beds significantly.

Fish Lake, located in the northwest corner of Dane County (south central Wisconsin), is a 100-ha seepage lake with an average depth of 7.3 m and a maximum depth of 20 m. Fish Lake has an extensive littoral zone with dense Eurasian watermilfoil beds extending for 100 to 200 m, from nearshore to a depth of about 4 m, and covering approximately half of Fish Lake's surface area.

Background and Objectives

Invertebrate abundance and community composition may differ dramatically among macrophyte species (Krecker 1939; Berg 1949; Krull 1970; Soszka 1975a; Gerrish and Bristow 1979; Menzie 1980; Dvorak and Best 1982; Cyr and Downing 1988a, b) or communities. Many of these differences have been attributed to differences in the structural morphometry or growth form of the plants.

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Plants with finely dissected leaf structure and/or greater surface area are generally thought to support more invertebrates (Berg 1949). Consequently, the manipulation of macrophyte communities by harvesting can affect the invertebrate food resources of fish, (Judd 1973; Schramm and Jirka 1989b), either directly by influencing invertebrate abundance and taxa composition (Poe, Schloesser, and Brown 1983; Rabe and Gibson 1984) or indirectly by influencing access to or availability of food resources (Pardue 1973; Diggins, Summerfelt and Mnich 1979; Diehl 1988; Savino and Stein 1982, 1989).

As the interest in macroinvertebrate-macrophyte associations has increased in recent years, much attention has been given to the exotic macrophyte Eurasian watermilfoil (*Myriophyllum spicatum* L.) and its associated macroinvertebrates (Balciunas 1982; Pardue and Webb 1985; Schramm and Jirka 1989a; Talbot and Ward 1987). The value of *M. spicatum* as habitat for macroinvertebrates (Menzie 1980; Miller et al. 1989; Pardue and Webb 1985) and fish (Keast 1984) has been investigated, but results of these studies are not conclusive. While some studies have demonstrated that milfoil provides excellent structural habitat for the attachment of aquatic invertebrates, other studies suggest that dense growths of plants inhibit the access of fish to these food resources.

Creating deep channels through these beds will increase the edge for attachment of invertebrates, and at the same time will provide greater access to predators. It is hypothesized that, by increasing the edge, we will increase predator-prey encounters (Smith 1993) which should, over time, improve the structure of predator populations through increased availability of prey, and improve the structure of prey populations by reducing the number of small fish in those populations. The Fish Lake study is also expected to provide information on milfoil's response to cutting and the increased utilization of invertebrate food organisms by smaller fish (Diggins, Summerfelt, and Mnich 1979; Fairchild 1982; Keast 1984; Mittelbach 1988; Schramm and Jirka 1989b). Cutting should decrease the total area of mil-

foil coverage but increase the amount of edge habitat and, consequently, the abundance of invertebrate food resources. Harvesting of watermilfoil also may stimulate the regrowth of native macrophyte species.

Results and Discussion

Three years of macrophyte data indicate that over 90 percent of the total macrophyte biomass of Fish Lake is Eurasian watermilfoil, which dominates the littoral zone with densities considered dense or even extremely dense (Figure 1). It is felt that the presence of these dense monotypic plant beds has influenced the biotic community in such a way as to produce a less desirable (i.e., stunted) panfish population and reduced the ability of predators to naturally control the panfish population. Engel (1985) observed that, while littoral zones support large numbers of fish, macrophyte beds act as screens, restricting fish movement, and that dense beds ($>300 \text{ g/m}^2$ dry weight) "were usually devoid of fishes...." Engel further observed that large bass were seldom found in the macrophyte beds until "die-back" formed channels through which the bass could cruise in search of food.

The Fish Lake largemouth bass population reflects this inability of bass to move through dense plants; diet analysis during summer (Figure 2) reveals that largemouth bass under 200 mm total length are consuming mostly insects, with some cyprinids, and darters. Whereas, bass longer than 200-mm total length switch to a diet of centrarchids. The switch from benthivory to piscivory is considered an important life history event and a major determinant in growth and fitness of fish, and has been documented to occur as early as 75-mm total length (Olson 1993) for largemouth bass from small Michigan lakes.

Relative weight is a measurement of condition or fitness based on the standard weight of a bass for each size class, where 100 is an optimum and fish falling below this level are seen as unfit. The relative weight of the bass appears to be closely related to this switch (Figure 2), as the relative weight increases to 100 after complete piscivory is achieved.

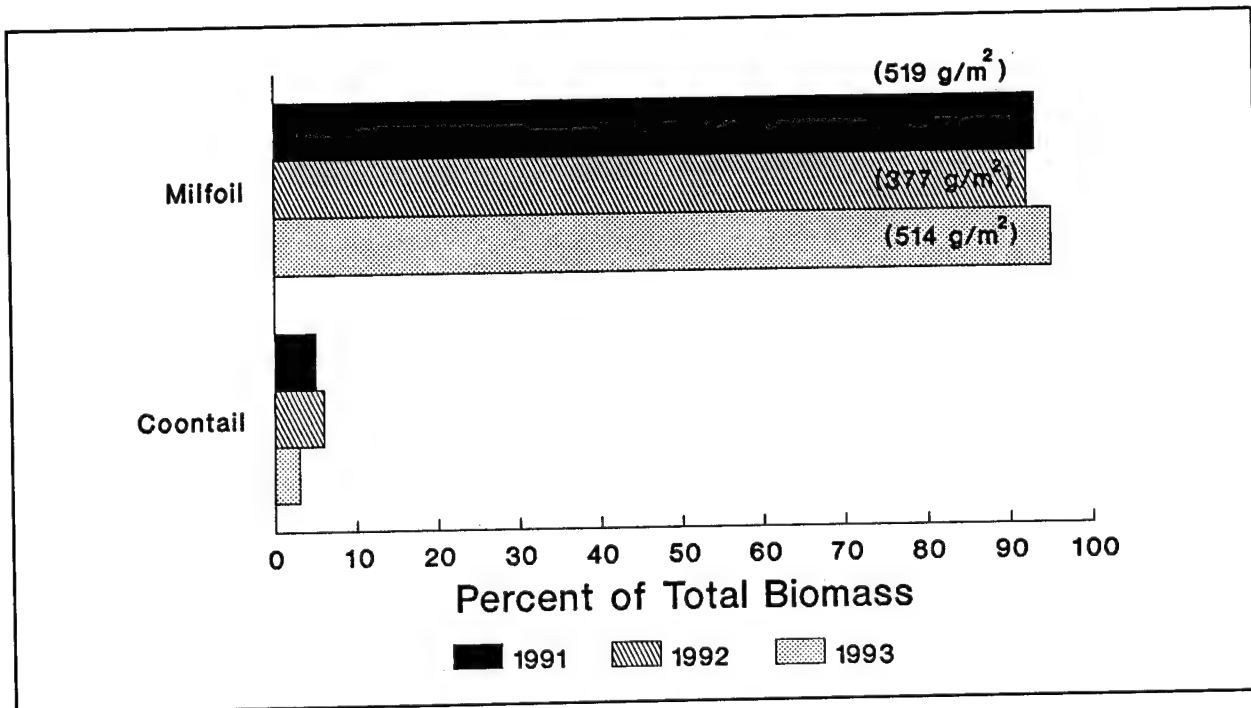


Figure 1. Fish Lake macrophyte relative biomass with actual Eurasian watermilfoil biomass shown in parentheses

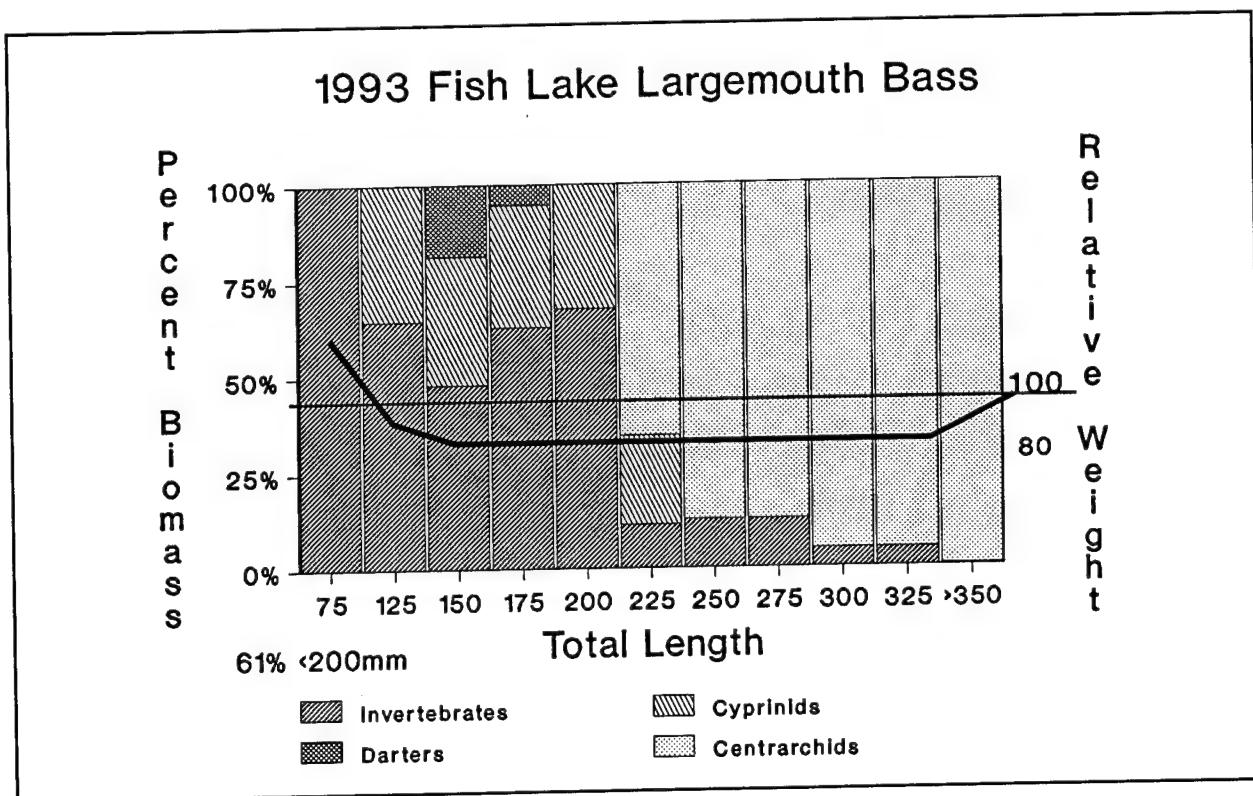


Figure 2. Largemouth bass diets by relative biomass, with relative weight shown on the right y-axis

Thus, it appears that the dense plant beds limit the ability of bass to capture centrarchid prey, such as bluegill and pumpkinseed.

Observations by SCUBA divers on Fish Lake indicate that bluegill and other prey species are most frequently found at the ecotone between dense macrophyte beds and open water areas (pers. observ.). Other researchers (Keast 1984; Werner, Mittelbach, and Hall 1981) have also reported congregations of bluegill near vegetation during the day. Bluegill choose to remain near or in vegetation where predators have difficulties capturing prey, thus leading to a large number of small bluegill (stunting) due to the inability of the predator to control the prey population. The low relative weight for all bluegill above 80-mm total length (Figure 3) is one indication that this is a truly stunted population wherein there are many small, thin fish. Length-frequency of bluegill captured in a purse seine versus electrofishing is shown in Figure 3 in percent to emphasize the point that the pelagic zone (purse seine) contains more large blue-

gill than in the littoral zone (electrofishing). The relative distribution of the population will probably follow the electrofishing distribution rather than the purse seine distribution, due to the larger sample size. Schnabel mark-recapture population estimates for Fish Lake bluegill indicate an abundance of small fish, with about 200,000 between 100 mm to 150 mm out of a total of approximately 220,000 fish (± 10 percent).

Bluegill diets (Figure 4a) reflect the distribution of invertebrates on plants (Figure 4b) in terms of percent occurrence. By percent occurrence bluegill diets and the plant-invertebrate distribution are dominated by zooplankton, chironimids, and mites, respectively, during May, July, and September (Figure 4). This indicates that bluegill are feeding in the littoral zone and are presented with two feeding choices in this habitat; one in the center of the plant bed where there is low predation risk and high competition for food and one at the edge of the milfoil bed where there is high predation risk and low competition for food.

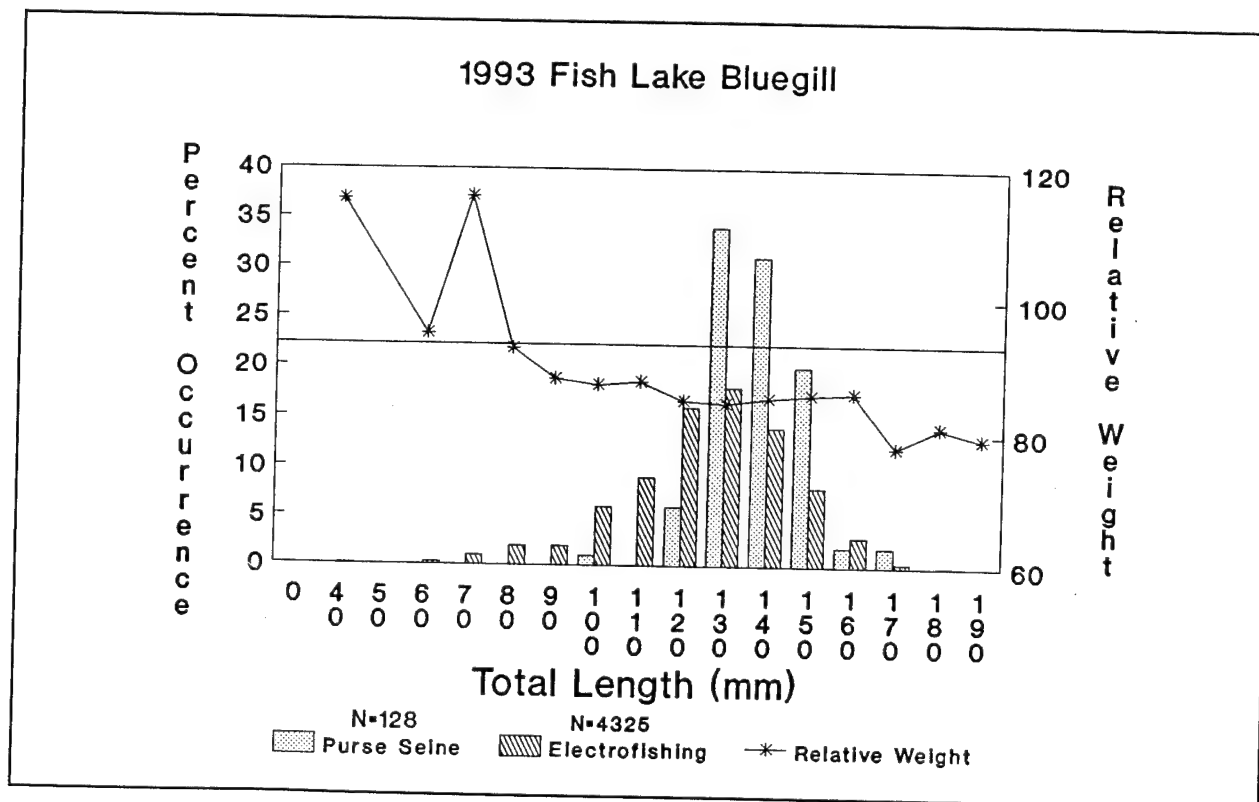


Figure 3. Bluegill length distribution by percent occurrence for purse seine and electrofishing gears. Relative weight is shown at the right

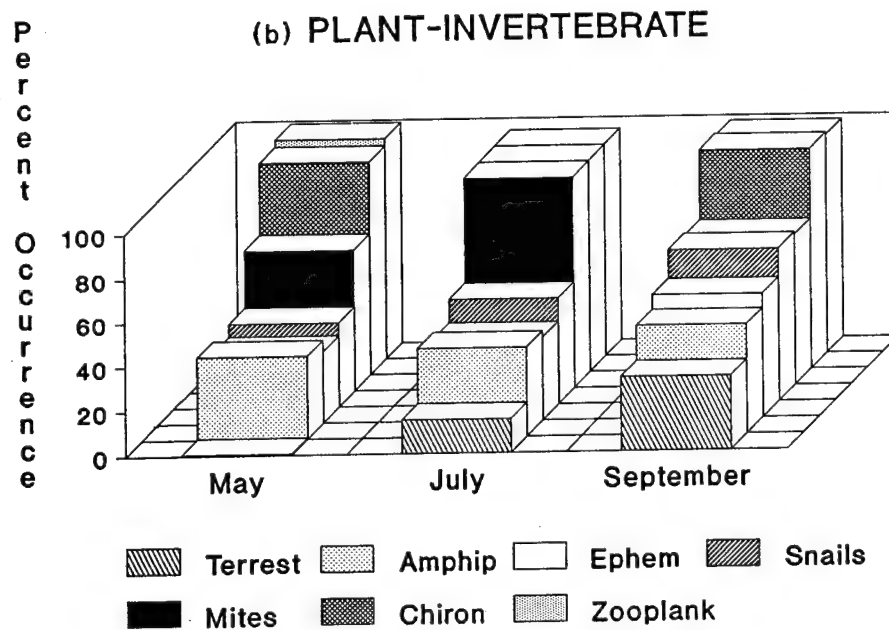
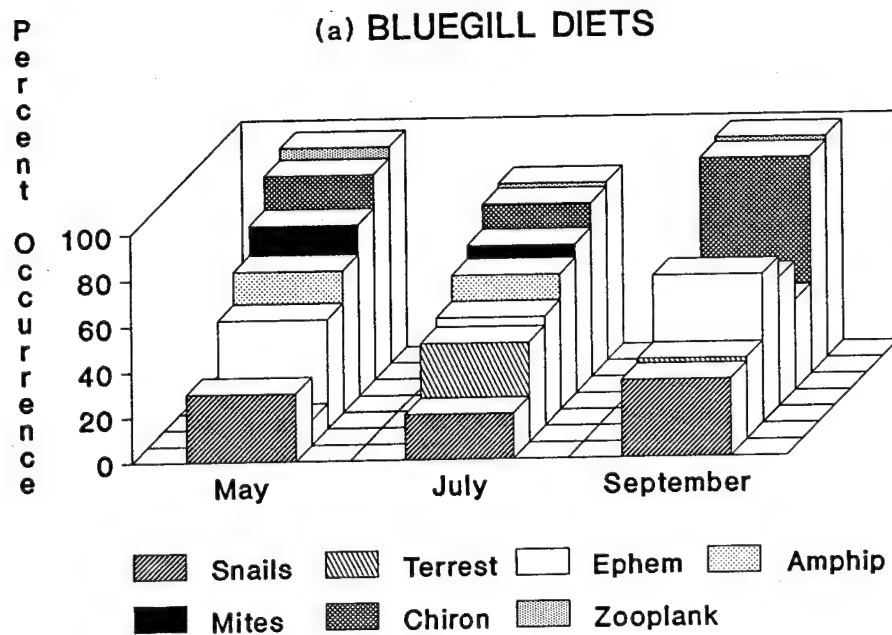


Figure 4. 1993 Fish Lake invertebrates by percent occurrence for (a) bluegill diets and (b) Eurasian watermilfoil

In addition to there being higher competition for food in the center of the milfoil beds, there may also be fewer invertebrate resources available due to an important aspect

of the growth form of *M. spicatum*; the plant progressively drops its lower leaves throughout the summer months. This process of sloughing is believed to result, in part, from

shading by the dense canopy cover of the upper shoots and leaves. The lower portions of plant stems in the interior of dense milfoil beds are mostly bare, while only the very upper portions are densely foliated (Madsen and Boylen 1990). This sloughing causes there to be fewer leaves and, therefore, less surface area for invertebrate attachment.

Plants located along the margins or edges of milfoil beds generally do not exhibit sloughing; or if they do, the length of unfoliated stem is generally not as great (Lillie and Budd 1992). If macroinvertebrate densities are indeed correlated with macrophyte biomass or surface area as suspected, then macroinvertebrate densities should be highest at the edges of milfoil beds. Our preliminary evidence supports this "edge" theory; for example, the total invertebrate biomass was significantly higher at the inner and outer edges than the center of the milfoil beds (Figure 5). Also, note that the apical and foliated sections of the milfoil supported more invertebrate biomass than the unfoliated sections of stem (Figure 5). Additionally, the inner edge supported a greater

total number of invertebrates (Figure 6) and number of species (Figure 7). These figures are based on log transformed data due to the fact that the original data were not normally distributed. This evidence suggests that the edge is a highly productive area for invertebrates. Thus, harvesting plant beds to increase edge habitat should increase food resources for fish, in addition to increasing predator-prey interactions. Additionally, some native plant species may recolonize the channel areas and have beneficial effects on littoral zone community dynamics.

Implications

Fish population dynamics are closely linked to macrophytes and associated invertebrates, thus littoral zone plant structure and abundance are important to fisheries. The density and species composition of macrophytes influence the distribution and abundance of macroinvertebrates, which serve as a food source for fish. Fish behavior is also influenced by the structure and density of macrophyte beds, affecting a

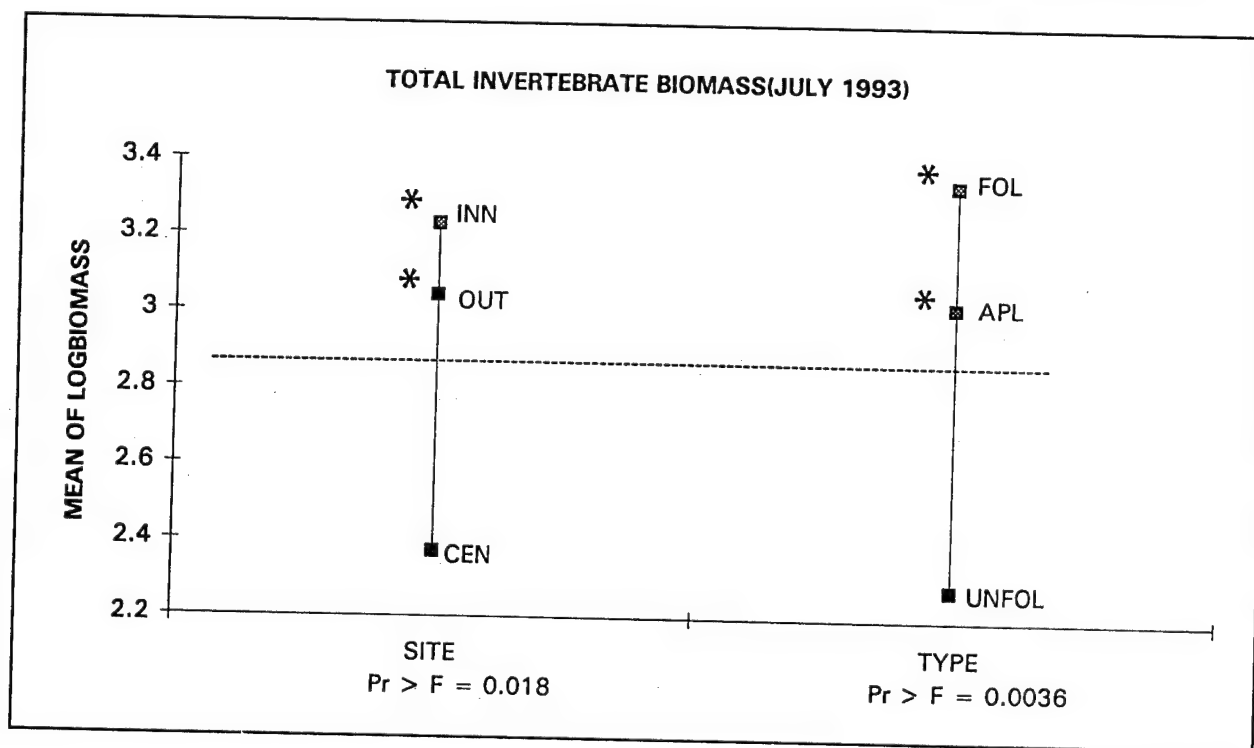


Figure 5. 1993 Fish Lake invertebrate distribution in the Eurasian watermilfoil beds by biomass, where the asterisk indicates that the mean log of the biomass is significantly higher at that site than at the lower site

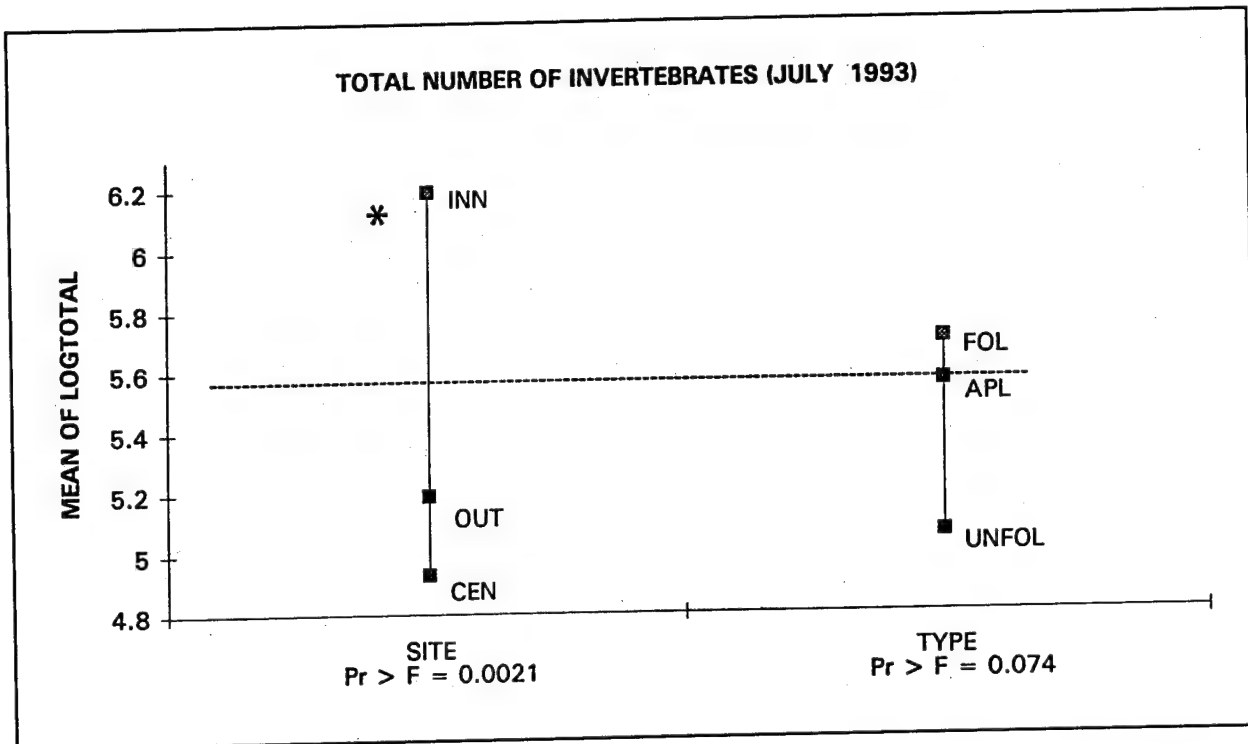


Figure 6. 1993 Fish Lake invertebrate distribution in the Eurasian watermilfoil beds by total number of invertebrates, where the asterisk indicates that the mean log of the total number of invertebrates is significantly higher at that site than at the lower site

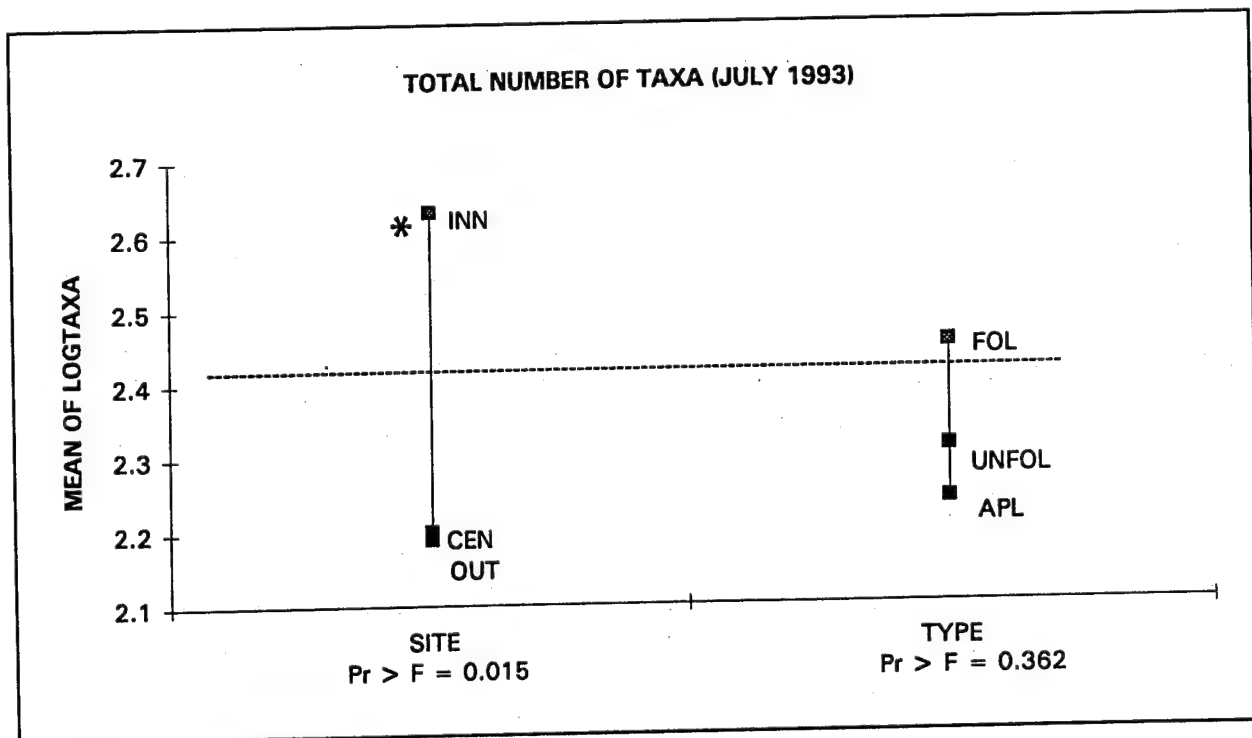


Figure 7. 1993 Fish Lake invertebrate distribution in the Eurasian watermilfoil beds by total number of taxa, where the asterisk indicates that the mean log of the taxa is significantly higher at that site than at the lower site

predator's ability to capture prey. Therefore, it is important for macrophyte management techniques to consider fish community effects.

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Effects of Stem and Leaf Complexity in Two Aquatic Plants (*Myriophyllum spicatum* and *Potamogeton nodosus*) on the Diets of Adult Largemouth Bass (*Micropterus salmoides*)

by

Eric D. Dibble¹ and Sherry L. Harrel¹

Introduction

Spatial complexity of aquatic plants creates habitat for many species of fishes (Cooper and Crowder 1979; Crowder and Cooper 1979; Diehl 1988; Crowder and Cooper 1982; Anderson 1984; Bettoli, Noble, and Betsill 1992; Killgore, Dibble, and Hoover 1993). Stem and leaves are a refugia from predators while providing substrate for the attachment of invertebrates that are fed upon by fishes (Pardue 1973; Savino and Stein 1982; Keast 1984). Factors affecting availability of food and the amount of spatial complexity in aquatic habitats are important because they affect growth, survival, and recruitment of freshwater fishes (Mittelbach 1981; Gutreuter and Anderson 1985; Mittelbach and Chesson 1987; Savino and Stein 1992).

Availability of food resources depends on plant architecture which may cause fishes to use aquatic habitats differently (Lillie and Budd 1992). Aquatic plant species vary in morphology, and differences in stem and leaf complexity alter foraging rates by decreasing capture rates and increasing searching times by fishes (Dionne and Folt 1991; Anderson 1984; Diehl 1988). Previous laboratory experiments using simulated aquatic plants suggest foraging efficiency by fishes decreases when the spatial complexity of aquatic plants increases (Savino and Stein 1982).

Evaluation of factors affecting fish distribution and foraging efficiencies in aquatic plants is important to understand how plant architecture influences growth and survival. Even

though previous experiments suggest aquatic plant complexity is important to foraging fishes, mechanisms affecting fish distribution within and among different aquatic plants have not been thoroughly studied.

In this experiment, differences between leaf and stem complexity are measured in two aquatic plants, and an enclosure experiment is conducted in a pond to investigate if differences of stem and leaf complexity in aquatic plants affect foraging behaviors of adult largemouth bass.

Methods

Experimental design

The experiment was conducted in a 0.5-acre pond at the LAERF, Lewisville, TX. The pond was lined with gravel (<25 mm in diameter) and a plastic barrier to prevent incidental growth of aquatic plants. Two plant species (*Potamogeton nodosus* and *Myriophyllum spicatum*) were planted and cultured in small plastic pools within each enclosure and replicated three times (Figure 1). Enclosures (approximately 10 m in diameter) were constructed with PVC pipe and plastic shade cloth (mesh size <0.5 mm).

Similar sizes and identical numbers of small prey fishes (100), 75 juvenile bluegill (*Lepomis macrochirus*) (\bar{X} = 69.7 mm TL) and 25 juvenile largemouth bass (*Micropterus salmoides*) (\bar{X} = 79.7 mm TL), were introduced into each enclosure (Table 1). After a 3-day acclimation period for prey fishes and approximately

¹ U.S. Army Engineer Waterways Experiment Station Vicksburg, MS.

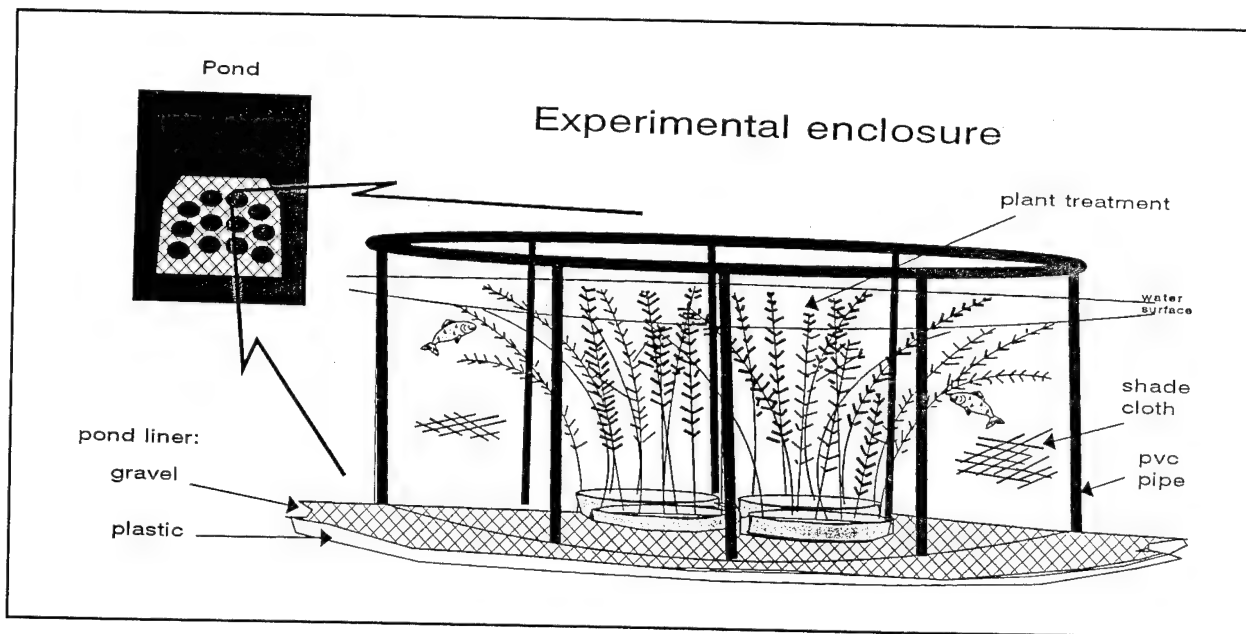


Figure 1. Experimental design of enclosures within the pond at LAERF

Table 1
Subsample Representing Sizes of Prey Fishes Introduced Into Experimental Enclosures

Juvenile Largemouth Bass			Juvenile Bluegill		
TL (mm) ¹	GTH (mm) ²	WGHT (g)	TL (mm)	GTH (mm)	WGHT (g)
74.0	16.5	3.94	73.0	23.0	4.96
78.0	16.0	4.03	66.0	22.0	3.87
65.0	14.0	2.58	75.0	25.0	6.01
93.0	19.5	7.27	61.0	19.0	2.83
68.0	14.0	2.96	69.0	23.0	4.38
82.0	18.0	5.50	76.0	24.0	5.47
95.0	20.0	8.22	66.5	20.0	3.60
68.0	15.0	3.07	71.5	22.0	4.56
93.5	19.5	8.02	66.7	19.5	3.78
80.5	17.0	4.50	72.0	22.0	4.50
$\bar{X} = 79.7$	17.0	5.00	69.7	22.0	4.40

¹ Total lengths of fish.

² Represents girth of fish; measurements taken from dorsal to abdominal portion of fish.

12 hr prior to introduction of predator fish, macroinvertebrates were sampled to determine prey availability. Dip-nets and plant removal were used to sample invertebrates. Three net

samples and three plant removals were collected in each enclosure, for a total of 18 invertebrate samples for each treatment. Net samples were taken by placing the net on the

bottom substrate and retrieving it quickly through the plants to the surface. Plant removal was accomplished by uprooting entire plants from substrate to surface and placing in bags to transport invertebrates to laboratories. All samples were preserved in 15 percent ethanol and later processed by identifying and recording abundances. Availability of invertebrates was expressed as percent relative abundance.

Five adult largemouth bass, similar in size ($\bar{X} = 367.9$ mm TL) were introduced into each enclosure (Table 2), for a total of 15 predator fish per treatment. Prior to introduction, all predators were kept approximately 7 days in aerated fiberglass holding tanks (approximately 3 m in diameter), so stomach contents would completely digest. After being introduced into

enclosures, predators were allowed to feed for approximately 48 hr and later retrieved by draining water from the pond. Diets were measured by removing and counting food items from stomachs of all predators. Prevalence of food items were expressed as percent occurrence and mean number per stomach.

Stem/leaf complexity

Complexity was measured along vertical and horizontal transects at three strata (upper, middle, lower). Horizontal transects (1 m in length) in upper strata of plants were taken within 150 mm from the surface, lower transects were placed approximately 300 mm above the substrate, and midlevel transects were centered between these measurements at

Table 2
Sizes of Predator Largemouth Bass Introduced Into Enclosures with Plant Treatments

<i>Potamogeton nodosus</i>				<i>Myriophyllum spicatum</i>			
Encl No. ¹	TL (mm)	GAPE (mm) ²	WGHT (g)	Encl No.	TL (mm)	GAPE (mm)	WGHT (g)
3	505.0	82.0	1988.9	4	435.0	74.0	1236.7
3	359.0	68.0	838.0	4	347.5	55.0	585.6
3	320.0	57.0	428.0	4	325.0	52.0	389.7
3	384.0	71.0	977.5	4	285.5	60.0	490.5
3	330.5	62.0	524.8	4	370.5	80.8	750.0
8	405.0	72.5	904.0	5	450.0	80.0	1278.9
8	370.0	65.5	674.6	5	359.0	66.8	820.5
8	370.0	60.0	695.8	5	552.5	54.5	541.5
8	365.5	66.0	665.0	5	290.5	52.0	385.8
8	324.5	56.5	489.0	5	338.0	60.5	567.8
9	385.0	63.5	737.0	7	445.5	73.0	1278.2
9	365.0	62.0	630.2	7	412.5	66.0	1023.2
9	355.0	62.0	549.5	7	345.0	58.5	514.8
9	340.5	56.5	605.9	7	295.9	53.5	405.0
9	285.0	53.0	389.0	7	320.0	55.0	448.5
$\bar{X} =$	364.3	63.8	739.8		371.5	61.3	711.9

¹ Enclosure number (three replicates per plant treatment).

² Gape measurements represent maximum distance between mandible and maxilla when mouth is open.

a depth of 1 m. Total length of transects for each plant replicate was 9 m, for a total of 27 m in each plant treatment. Vertical transects (0.5 m in length) were run from the surface down, from the substrate up, and centered between these lower and upper measurements at a depth of approximately 1 m. Average length of vertical transects in each enclosure was 4.5 m, for a total of 13.5 m in each plant treatment. Size and abundance of interstices (gaps between stems and leaves) were measured along transects with a point intercept method. Plant complexity was determined similar to Lillie and Budd's (1992) architectural index, and Dibble's gap analysis (Dibble and Killgore 1994), where the sum of horizontal and vertical interstice ratios (I_{hv}) were determined. Ratios (I_i) represented the number of interstices (gaps between stems and leaves) intercepted per meter (r_i) and the mean size (cm) of each interstice (x_i). Values were expressed as *stem/leaf-complexity*. Significant differences between treatments in invertebrate availability, stem/leaf-complexity, and diets were tested with a one-way AOV (Statistix 1994).

Results

Treatment effect on fish diet

A total of 126 invertebrates and 72 prey fishes were removed from adult bass. Significant differences ($F = 20.49$, $P < 0.001$) in diets were noted between plant treatments. Invertebrates were the primary diet of predators foraging within enclosures containing *P. nodosus* (Figure 2). A mean of 83.6 percent invertebrates and 16.4 percent fish were recorded across replicated samples from fish in this plant treatment (Figure 2). However, more fish than invertebrates were consumed in *M. spicatum*. Invertebrates constituted only 16.7 percent of stomach contents across replicates, while 83.3 percent of the diet were prey fishes (Figure 2). Juvenile bluegill and largemouth bass were the most abundant food items from bass foraging in *M. spicatum*, 5 fish/stomach and 2.33 fish/stomach, respectively (Figure 3). Two families of dragonfly larvae, Aeshnidae and Libellulidae, were the most common invertebrates consumed in

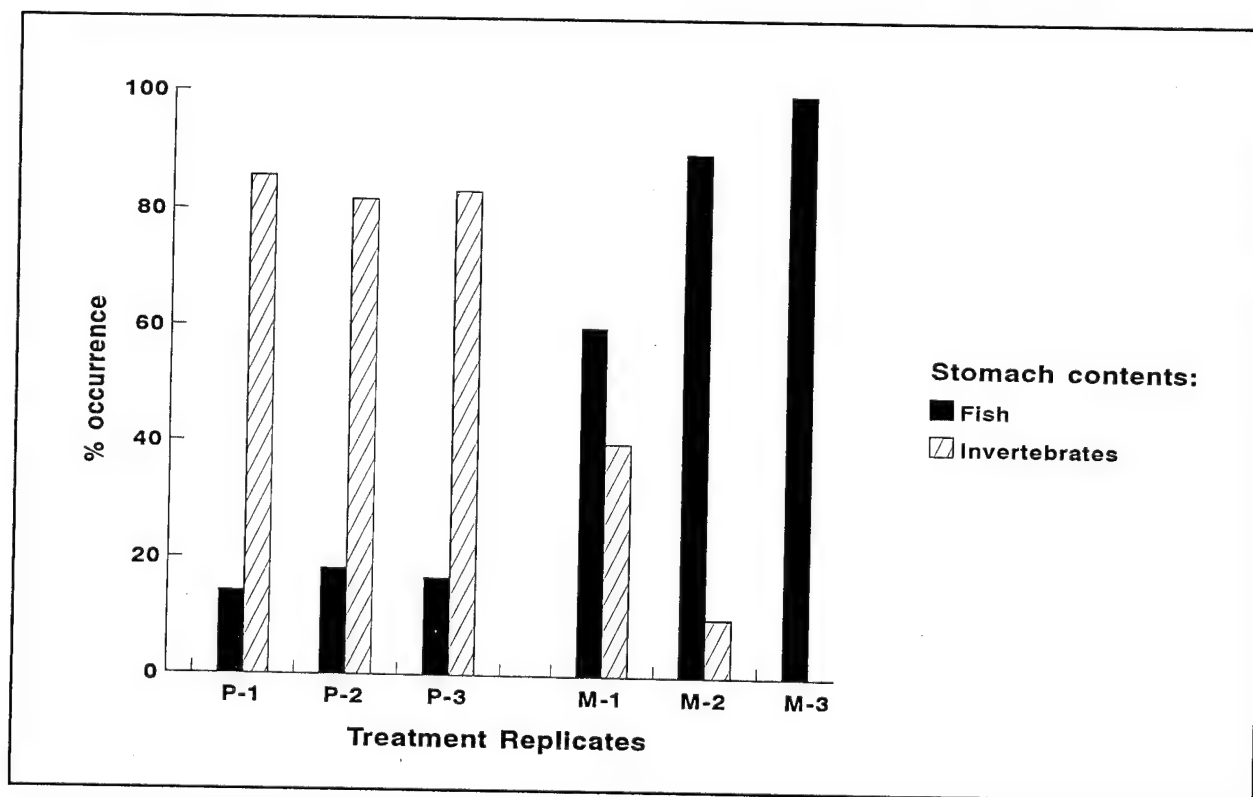


Figure 2. Treatment differences in the prevalence of fish and invertebrates in the diets of adult largemouth bass

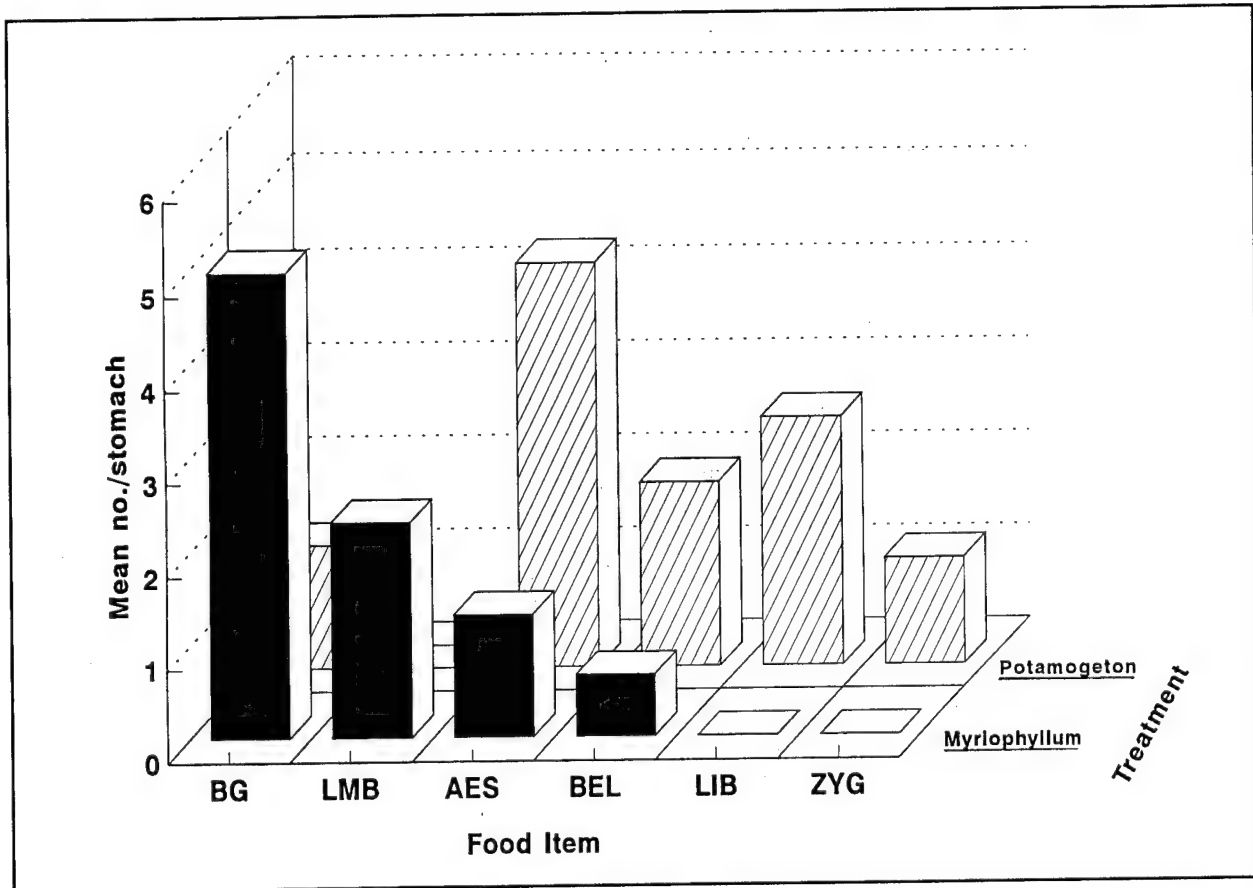


Figure 3. Treatment differences of stomach contents in the predators of specific food items (BG = juvenile bluegill; LMB = juvenile largemouth bass; AES = Aeshnidae; BEL = Belostomatidae; LIB = Libellulidae; and ZYG = Zygoptera)

P. nodosus, 4.33/stomach and 2.67/stomach, respectively.

Stem/leaf complexity

Significant differences ($F = 17.80$, $P < 0.01$) were noted in stem/leaf complexity between plant treatments (Figure 4). Mean complexity of *P. nodosus* was high ($\bar{X} = 3.29$) compared to *M. spicatum* ($\bar{X} = 1.3$). Stem/leaf complexity varied across plant strata more in *M. spicatum* than in *P. nodosus*. Lower, middle, and upper stem/leaf complexity of *P. nodosus* were similar, 3.25, 3.0, and 3.62, respectively. Stems and leaves were most complex in *M. spicatum* at lower strata than middle and upper strata (Figure 4).

Invertebrate availability

No significant difference ($F = 0.08$, $P > 0.5$) was noted in the relative abundance of large

invertebrates between treatments. Dragonfly larvae (Libellulidae, Zygoptera) represented most of the invertebrates sampled in both plant treatments (Figure 5). Libellulidae made up over 50 percent and Zygoptera over 25 percent. Aeshinidae represented approximately 11 percent of the sample in both treatments, and Belostomatidae (water beetle) a little over 3 percent.

Discussion

Results from this experiment suggest that aquatic plant morphology (i.e., stem/leaf complexity) significantly influences foraging behaviors of adult largemouth bass. Food items were numerically equal across plant treatments yet differences in predator diet were observed, suggesting that differences in stem/leaf complexity were a causative factor in foraging success. Decreases of larger mobile prey (i.e., juvenile fishes) and increases of smaller

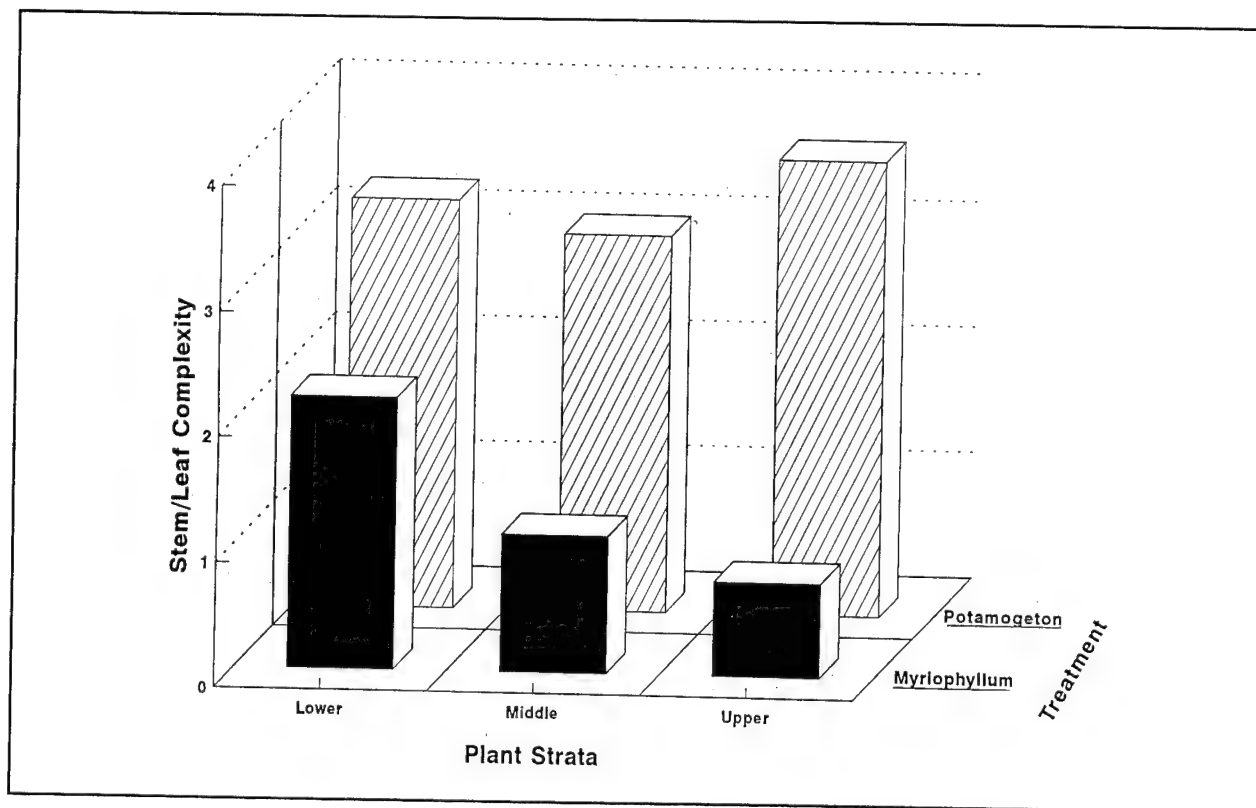


Figure 4. Treatment and plant strata differences in stem/leaf-complexity

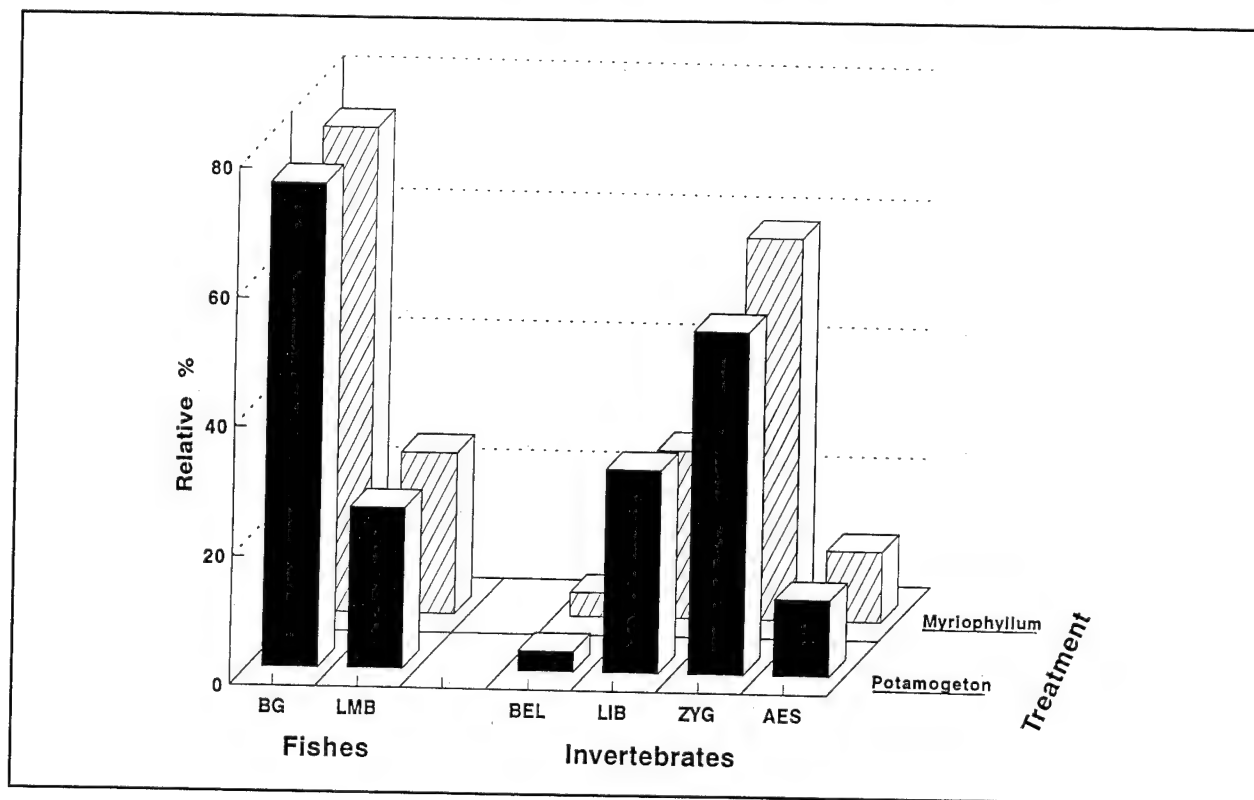


Figure 5. Relative percent of prey items within treatments. Fishes were introduced, and invertebrate data represent samples collected within plants in enclosures and across treatments

less-mobile prey (i.e., invertebrates) in diets of fish foraging in *P. nodosus* with higher stem/leaf complexity were noted. Inversely, diets of fish foraging in the plants of low complexity (*M. spicatum*) showed fewer juvenile fishes but higher numbers of invertebrates.

These results agree with others that showed foraging efficiency decreases when spatial complexity increases, and that interference of stems and leaves do decrease capture success in foraging fish (Mittlebach 1981; Savino and Stein 1982; Anderson 1984; Diehl 1988). Apparently, *P. nodosus* decreased capture success of larger, more mobile juvenile fishes, and predators switched to alternative invertebrate prey that were less mobile and easier to capture. However, foraging success for prey fishes in *M. spicatum* was high because of low stem/leaf complexity.

Efficiency at which fishes obtain their prey is important, because of the tradeoff between energy requirements for food intake and energy allocated for growth (Anderson 1984; Diehl 1988). Growth is important to fish survival because predator risk and mortality are frequently size dependent (Gutreuter and Anderson 1985; Adams and DeAngelis 1987; Sih 1987). Fish spending additional time searching for food or obtaining smaller prey expend extra energy in foraging, and thus less energy is allotted toward growth (Mittlebach 1981; Gutreuter and Anderson 1985; Adams and DeAngelis 1987). Fish foraging in habitat high in spatial complexity are thus less likely to grow as large as fish foraging in habitat with low spatial complexity with the same food resource.

If the assumption is made that foraging efficiency depends on amount of spatial complexity in aquatic habitats, it is reasonable to conclude that aquatic plant architecture and morphology (i.e., stem/leaf complexity) are important for determining the quality of aquatic habitats. This research represents a preliminary step in classifying and evaluating important aquatic plant criteria which benefit growth and survival of fishes. Future study is required, however, before a more thorough understanding

of how differences in plant morphology and architecture influence freshwater fishes.

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Simulation Technology

Simulation Technology Team Overview

by
John D. Madsen¹

The purpose of the Simulation Technology Team is to develop and produce computer tools which assist in planning for aquatic plant management. These tools may consist of simulations of plant growth or response to management techniques, techniques to integrate Geographic Information Systems (GIS) into aquatic plant management planning, or access to information required to develop management plans.

The current work units include plant growth models, biological control models, chemical control models, GIS and plant management database development, and the aquatic plant management strategy planner (Table 1).

Plant Growth Models

The plant growth model work unit focuses on the development of PC-based plant growth simulations of the three major nuisance plants: waterhyacinth, Eurasian watermilfoil, and hydrilla. Models of these three species would not only be used to predict potential nuisance levels in water bodies under varying environmental conditions, but would also be incorporated into models of various control techniques. Therefore, this work unit is the basis of developing all simulation models for control techniques. Development of models for all three species has been progressing, with a model of Eurasian watermilfoil nearing release. In the future, generalized models for

Table 1
Work Units for FY94 Under the Simulation Technology Area

Work Unit Name	Work Unit Purpose	Products Under Development
Plant Growth Models (32440)	Develop simple growth models of waterhyacinth, Eurasian watermilfoil, and hydrilla. Models will be used in both predicting infestations and as plant response component in management technique models	HYACINTH HYDRILLA MILFOIL
Biocontrol Simulation (32438)	Develop simple models of the effect of biocontrol agents on target plants, such as insect biocontrol agents on waterhyacinth and grass carp on hydrilla and Eurasian watermilfoil	INSECT AMUR STOCK
Chemical Control Simulation (32439)	Develop computer tools to assist in chemical control operations and evaluation.	HERBICIDE
Database Procedures for Effective Aquatic Plant Control Methods (32506)	Develop computer-assisted data collection and analysis procedures to support operational plant control programs and simulation technologies	(GPS and GIS Procedures)
Development of an Aquatic Plant Management Strategy Planner (32954)	Develop an integrated computer-assisted aquatic plant management planning tool, incorporating information bases, simulations, and a planner	APROPOS

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native species may also be developed to provide insight into the growth patterns of desirable species under different management regimes.

Biological Control Models

The biological control model work unit will end after 1994. During the tenure of this work unit, models of the interaction of waterhyacinth with its biocontrol insects were developed and released. However, sufficient data to continue this type of modeling effort for other biological control agents on target species were not available. Therefore, revisiting this type of simulation effort on biocontrol agents for Eurasian watermilfoil and hydrilla will require more information first.

As part of this work unit, a model to determine grass carp stocking rates (AMUR STOCK) was also developed. As with most of the simulations, it requires initialization data. However, it can give some insight into the results of different stocking rates on resulting plant management levels. AMUR STOCK will be released in a Windows[®] version in early 1995.

Chemical Control Simulations

The chemical control simulations work unit has focused on development of a herbicide dissipation simulation, HERBICIDE. HERBICIDE provides information on herbicide uptake and dissipation rates for plant, sediment, and water column based on known parameters. This information is helpful not only in establishing environmental residue information, but also can be used to determine water column concentrations and exposure times for use with known relationships between concentrations and exposure times for given compounds and plant species, as developed by the Chemical Control Technology Team. HERBICIDE will be released in a Windows[®] compatible format in early 1995. Continuing development will be a cooperative effort with both the Hydraulics Laboratory to focus on improving water movement parameters and the Chemical Control Technology Team to

validate the model against the extensive data from the triclopyr dissipation study on Lake Minnetonka.

GIS/Database Development

The database development work unit has focused on acquiring available plant management data, and integrating GIS data and tools with planning management strategies. Direct input of data such as maps derived from global positioning systems (GPS) have also been investigated. This work unit has also attempted to integrate the GIS data and visualization techniques with output from plant growth simulations and control simulations, such as AMUR STOCK. Ongoing research includes further integration of mapping techniques, and integrating ArcInfo[®] and ArcView[®] into an aquatic plant management strategy planner (see below).

Aquatic Plant Management Strategy Planner

In this new work unit, a technology transfer tool is being developed to integrate available simulation models, information on control techniques, databases, and instructional guides with a tool that will assist in developing systematic aquatic plant management plans. A demonstration interface, under the name AP-ROPOS (Aquatic Plant Resource Operations & Planning Online Support), has been developed for evaluation. An information exchange bulletin will be published soon on this system. In addition, a questionnaire on developing the planner will soon be available on the Aquatic Plant Bulletin Board System.

Other Models

The simulation model HARVEST, which evaluates harvesting operations, is currently being converted to a Windows[®] compatible format. This will greatly increase the ease with which this model can be used. In fact, conversion to a graphics user interface is planned for all of the Simulation Area models over a period of time. Using this format, all models will be accessible through APROPOS.

The recent establishment of the Aquatic Plant Bulletin Board System (APBBS) through the Center for Aquatic Plant Research and Technology (CAPRT) is an important tool for increased interaction and technology transfer. The APBBS will allow exchange of databases and upgrades of simulations. Questionnaires on the APBBS will allow for interaction and exchange of views on specific topics. The Simulation Technology Team will utilize the APBBS in model development and support.

With the initiation of APROPOS, a new direction for the Simulation Technology Team has begun. APROPOS will incorporate simulations as one important computer tool for aquatic plant management planning. The simulations will be fully accessible and integrated within that system. In addition, other tools and techniques will be integrated to form a single planning interface.

Aquatic Plant Growth Simulations

by
R. M. Stewart¹

Background

Natural variability in both temporal and spatial patterns of aquatic plant growth make planning effective, yet efficient, control strategies difficult for aquatic plant managers. To a large part, this variability in plant growth is due to seasonal and spatial differences in environmental conditions. Currently, management strategies for nuisance aquatic plant infestations are often based on successes and failures from previous years. If the infestation is new, management decision-makers may rely on actions implemented at locations having similar infestations, site conditions, and management objectives. Though this method of management planning has resulted in numerous aquatic plant management successes, new situations often arise that have few similarities with other sites. Under these situations, managers have few tools to help with their planning efforts.

The Simulation Technology task area of the APCRP is responsible for development of PC-based simulation models for three problematic, exotic aquatic plant species. The plant species are waterhyacinth (*Eichhornia crassipes*), hydrilla (*Hydrilla verticillata*), and Eurasian watermilfoil (*Myriophyllum spicatum*). The simulations will function to provide plant growth information useful for evaluating the effects of generalized site conditions on seasonal growth of these plant species. Information resulting from these simulations will be used to evaluate the spatial and temporal growth patterns of these plant species at target sites. This type information will help aquatic plant managers plan optimal control strategies for these plant species based on site conditions and management objectives.

Model Considerations

What type information will the models provide?

The models are designed to generate daily estimates of plant biomass resulting from growth of a given plant species under user specified site conditions. An understanding of the effects of site conditions on seasonal plant growth patterns helps managers plan more effective control strategies for problem infestations. Information of the type illustrated in Figure 1 will help managers anticipate when and where plant biomass levels will attain problem levels. This capability will allow them to plan control measures that effectively prevent the occurrence of nuisance plant growth. Additionally, information generated by the models will provide a basis for evaluating effectiveness of control treatments. What type site conditions are considered? Each of the species specific plant growth models can be initialized for composite site conditions of daily temperatures and solar irradiance levels, daily photoperiod values, and initial plant biomass and tuber density (hydrilla only). For submersed species models, water depth and daily values for water temperature and water transparency are also requested. In their current form, these simulations assume that nutrients are not limiting. Because composite site conditions are heterogenous across locations within a water body, the models must be run separately for each unique set of site conditions (i.e. a single run of these models cannot be used to simulate growth of the respected plant species for all locations within a water body).

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

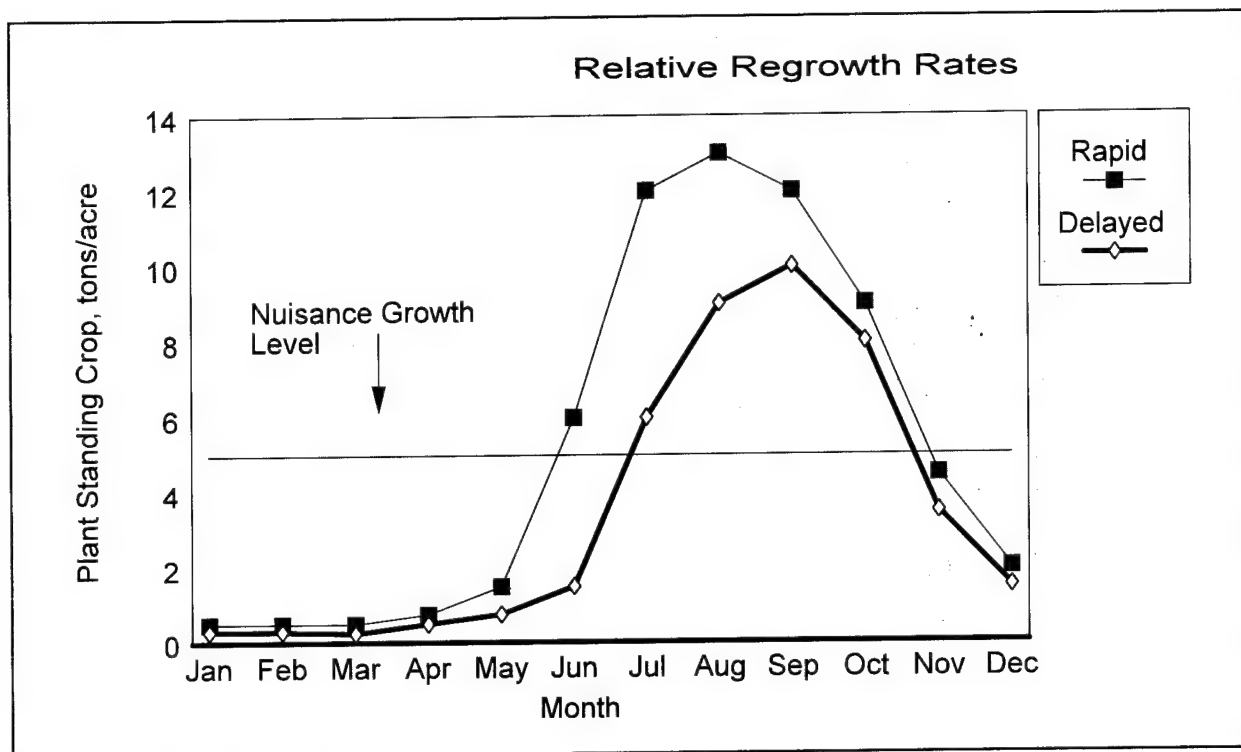


Figure 1. Example outputs of monthly plant biomass estimates generated by the models for two different locations in a reservoir

How will the models consider spatial variability?

Spatial variability in plant growth patterns within established colonies of the same species often result as a consequence of spatial differences in environmental conditions. For example, differences in Eurasian watermilfoil seasonal growth patterns would likely occur, even within an established colony, along a depth gradient within the colony. Differences in growth rates along a depth gradient are likely to occur due to the increase in the time required for developing shoots to elongate to the water surface. Typically in productive waters, biomass production is limited prior to the shoots nearing the water surface where light penetration is sufficient to support growth. Differences in growth patterns would also be expected between different species, even when the species are coexisting under the same environmental conditions.

As stated above, the models must be reinitialized and run for each unique combination of site conditions. As an aid to the extensive data management requirements, geographic in-

formation system (GIS) techniques are currently being used to help organize site classifications (Figure 2) based on environmental conditions and to assist in spatial visualization of model outputs (Figure 3).

How will this information help plan mechanical harvesting operations?

Mechanical harvesting operations consist of the cutting, collecting, and transporting (i.e. removal) of aquatic plant biomass. Productivity of both the collection and transport processes is extremely sensitive to the amount of harvestable biomass, as well as other operational parameters. The HARVEST simulation model was developed at WES to provide a tool capable of systematically evaluating mechanical control system performance without the need to conduct expensive field trials. Information generated by the plant growth models on seasonal biomass levels will help managers evaluate mechanical harvesting systems and plans. By including the spatial link to GIS-type databases, harvesting system evaluations can more easily consider both temporal and spatial variability in plant biomass.

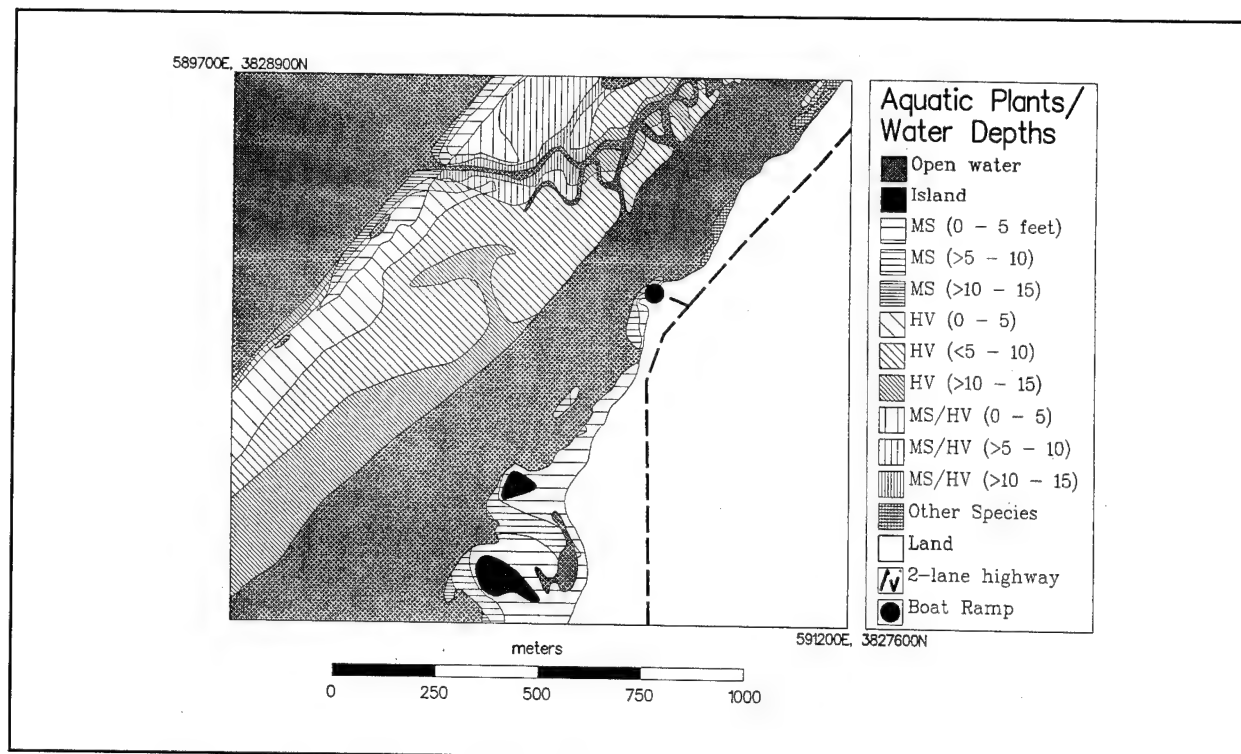


Figure 2. Classification of aquatic plant sites based on plant species and water depths

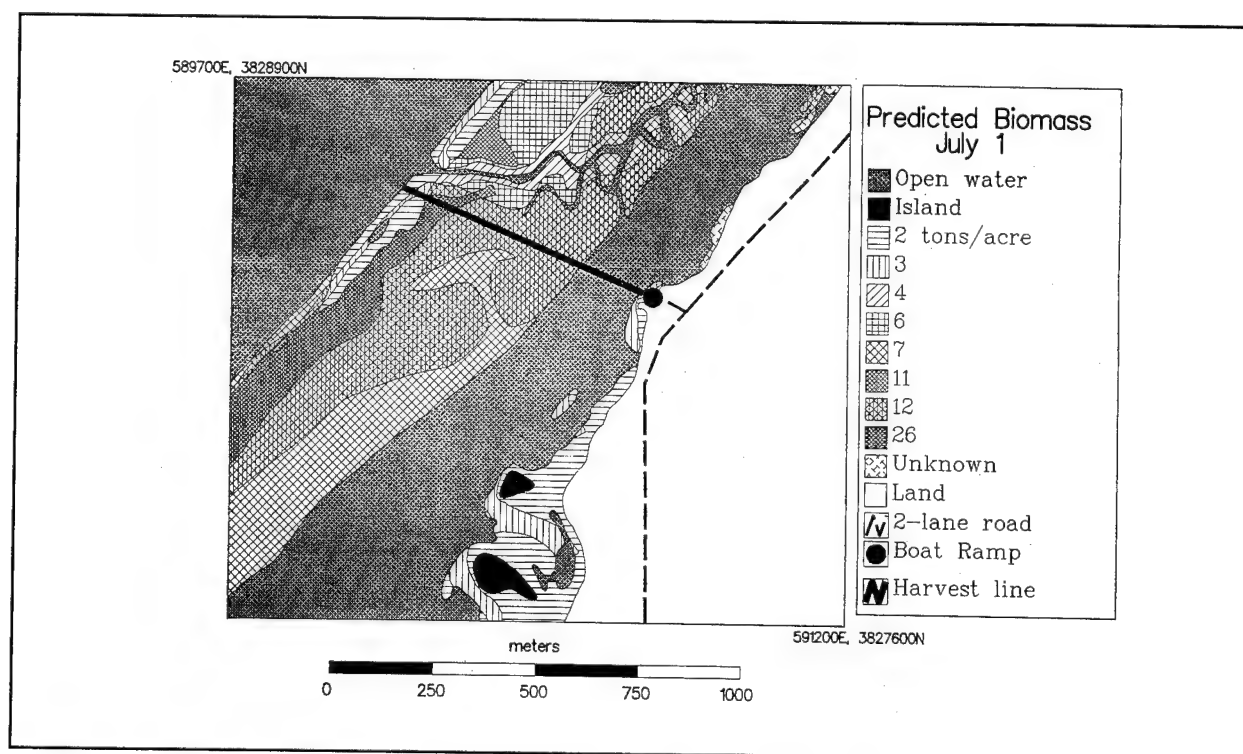


Figure 3. Spatial registration via GIS of "peak" plant biomass estimates generated by the plant growth models

Additionally, GIS techniques will also provide a method for obtaining additional HARVEST model initialization data, including areal dimensions of the harvest site and over water transport distances from the harvest site to the onshore disposal site.

How will this information assist in planning biological control strategies?

Successful control of aquatic plant growth by herbivorous grass carp requires that stocked fish consume more vegetation than is produced by plant growth. As described by Boyd and Stewart in this Proceedings (see pages 290-294), plant growth models are used in conjunction with a module for white amur feeding to generate estimates of stocking requirements for different target plant species growing under different sets of environmental conditions. Outputs generated by the WES AMUR/STOCK model, which incorporates simplified versions of the Eurasian watermilfoil and hydrilla growth models, indicate that stocking rate requirements are very sensitive to regrowth rates of overwintered plant populations and to peak biomass levels.

How will this information assist in planning chemical control strategies?

Unlike biological control and mechanical control systems, efficacy of chemical control techniques for submersed plants is not highly

sensitive to the standing crop or growth rate of the target vegetation. For herbicide applications to be effective, sufficient quantity of the active ingredient must be applied to the water column to attain the proper aqueous concentration and exposure time. For this reason, plant growth rates and biomass levels have relatively minor effects on herbicide effectiveness. Estimates of when and where plant growth will attain nuisance levels, however, can help resource managers develop application schedules that prevent nuisance growth levels from interfering with intended uses of the water body.

What is the current status of these plant growth models?

Work is continuing on the Eurasian watermilfoil and hydrilla plant growth models, with an initial release of the MILFOIL model scheduled for FY95. Even as these models near their initial releases, selected improvements and additions are being planned. For example, alternate techniques for predicting dispersal and establishment of submersed aquatic plants are currently being evaluated to assist in developing system-wide planning tools. These techniques will likely be based on regression techniques, as opposed to the processed based simulation models currently under development.

Biological Control Simulation - The AMUR/STOCK Model

by
W. A. Boyd¹ and R. M. Stewart¹

Introduction

Several biological methods are now available to control nuisance aquatic plants. Included among these methods is the use of herbivorous grass carp. Since their introduction into the United States in 1963 (Guillory and Gassaway 1978), grass carp have proven to be an effective means of aquatic plant control. Ecological concerns, however, arise because of the uncertainty in determining stocking rates to provide desired control levels without risking unwanted impacts (Noble, Bertolli, and Bestill 1986, Leslie et al. 1987). Variability in stocking rates that have been used under different conditions is exemplified in Table 1.

The purpose of this paper is to illustrate use of the White Amur Stocking Rate Model developed by the WES for evaluating the effect of factors which can contribute to variability in stocking strategies. While AMUR/STOCK can by no means consider all the bio-

logical and environmental factors, it does allow the user to examine the effects of user-defined variability for some of these factors. These factors can be divided into three general categories: (1) Site conditions, (2) Plant infestation levels, and (3) Grass carp.

General Initializations

Certain initialization conditions were constant for all simulation runs. These included the seasonal water temperature dataset which was taken from average median monthly water temperatures from Guntersville Reservoir, AL, for the 4-yr period 1984-1987. Average size of stocked fish was initialized at 0.34 kg, an approximation of the average weight of a 30-cm fish (Kirk, Morrow, and Killgore 1994). All simulations additionally considered that the target plant was *Hydrilla verticillata*, a plant that ranks highly on most feeding preference lists of grass carp (Sutton and Van Diver 1986; Leslie et al. 1987). Finally, grass carp

Table 1
Stocking Rate Requirements for Case Studies

Source	State	Stocking Rate
Sutton and Vandiver (1986)	Florida	3 - 638 fish/veg hectare
Leslie et al. (1987)	Florida	9 - 440 fish/veg hectare
TVA (1990)	Alabama	17 fish/veg hectare
Klussman et al. (1988)	Texas	74 fish/veg hectare
Santha et al. (1991)	Texas	79 - 130 fish/veg hectare
Wiley et al. (1987)	Illinois	47 - 370 fish/veg hectare
Bonar et al. (1993)	Oregon	180 fish/veg hectare
Stocker and Hagstrom (1985)	California	6,323 fish/veg hectare

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

losses resulting from off-target feeding and escaping were set to zero percent for all simulations generated.

Levels of Peak Biomass

Using general initializations described above, grass carp stocking requirements were determined for control of plant growth that attained peak biomass levels of 110 g/m^2 , 220 g/m^2 , and 440 g/m^2 . These biomass values equate on a fresh weight basis to approximately 11, 22, and 44 metric tons per hectare, respectively. In field situations, these differences in peak biomass could occur due to differences in colony age, or to environmental factors including water depth, light availability, and sediment nutrient levels.

For each peak biomass level, plant growth was initiated with an overwintering biomass level of 11 g/m^2 , or approximately 1 metric ton per hectare. The resulting plant growth curves, generated using different calibration datasets for the effects of season and temperature on growth rates, are shown in Figure 1. Note that the spring regrowth rates are similar for all curves, each having similar slopes during the early spring period.

Simulation outputs were generated to provide estimates of the stocking rates required to control plant growth for each peak biomass level (Figure 1) by late summer of poststocking Years 1-5. Stocking rate requirements were sensitive to the poststocking time. Stocking requirements ranged from 156 fish/ha for control of high peak biomass growth in Year 1 to 10 fish/ha in Year 5. On the other hand, stocking requirements were only slightly sensitive to peak biomass levels. Differences in the stocking requirements of the high and low peak biomass simulations were approximately 10 percent for each poststocking year.

Levels of Overwintering Biomass

Overwintering biomass levels can vary due to differences in plant species, geographic region, water-body types, and locations within a water body. Therefore, the effect that the overwintering biomass level can have on stocking rate requirements was evaluated with the model. Overwintering biomass levels considered were 11 g/m^2 , 44 g/m^2 , and 110 g/m^2 . The lower level could represent plant colonies

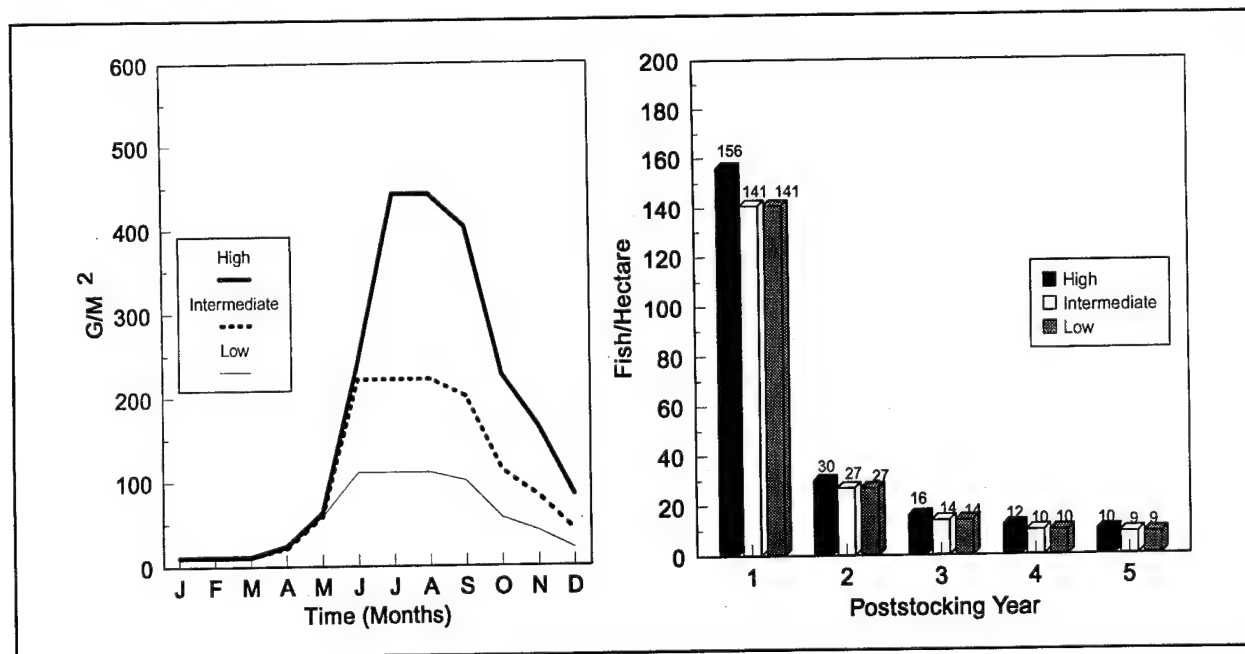


Figure 1. Seasonal plant biomass curves generated by the AMUR/STOCK model for different peak biomass levels (left) and their resultant effect on grass carp stocking rate requirements (right)

in which the majority of the shoot material generated during the growing season is lost through senescence or by other processes. The highest overwintering biomass level could represent plant colonies which overwinter intact.

Peak plant biomass for each of the simulations was initialized at 440 g/m^2 . Resulting plant growth curves are illustrated in Figure 2. Note that although regrowth rates are similar for each simulation, peak biomass was attained approximately two months earlier in the high overwintering biomass simulation.

Stocking rate requirements for the three overwintering plant biomass levels are shown in Figure 2. Stocking rate requirements were sensitive to both overwintering biomass levels as well as to poststocking time. Consistently throughout the 5-year simulations, the stocking rate requirement for the high overwintering biomass level more than doubled that of the intermediate biomass level and was approximately three times higher than that of the low biomass level.

Onset of Plant Regrowth

The onset of regrowth for plant colonies from overwintering tissues may vary according to a number of factors. These factors include plant species, water-body temperature, as well as the rooting depth of the plants. Timing of plant regrowth initiation was evaluated for its effect on grass carp stocking requirements by considering plants that initiate regrowth in April (REGULAR), plants that initiate regrowth in June (DELAYED UNTIL JUNE), and plants that initiate regrowth in July (DELAYED UNTIL JULY). For each of these three plant growth curves, plant overwintering biomass levels were initiated at 11 g/m^2 and plant peak biomass levels were set at 440 g/m^2 .

Resultant plant growth curves displayed in Figure 3 show the plants achieving the same peak biomass but during different months. During regular regrowth, plants are shown to reach their peak biomass during June. When

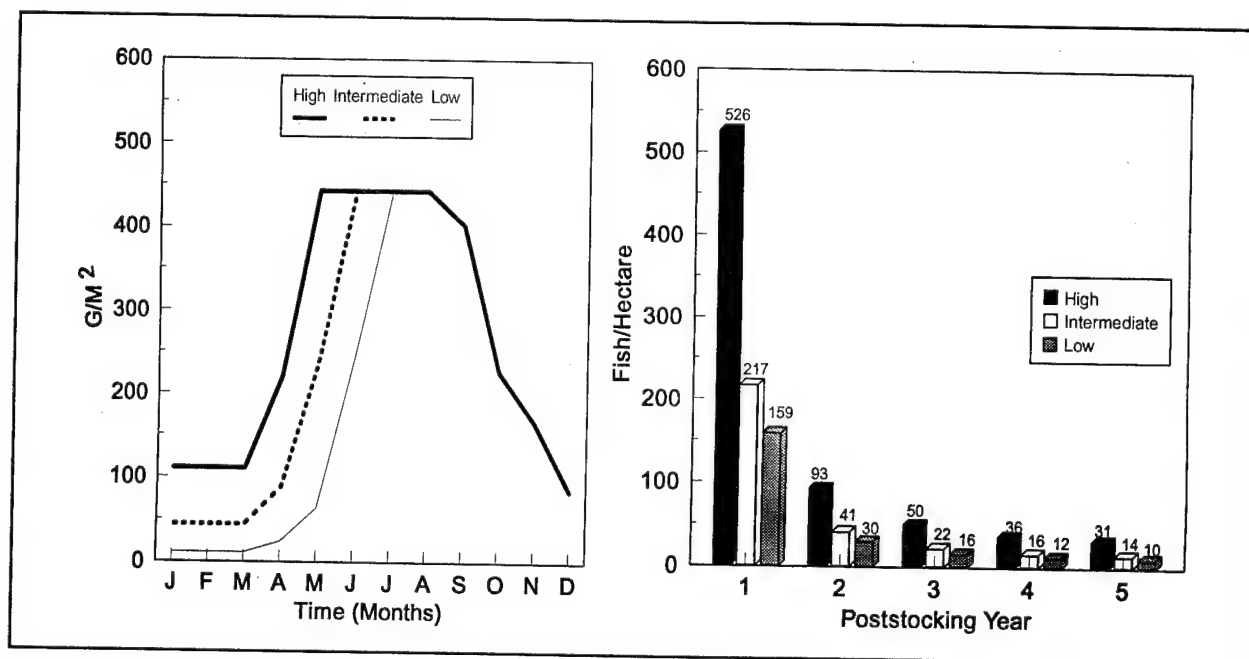


Figure 2. Seasonal plant biomass curves generated by the AMUR/STOCK model for different overwintering biomass levels (left) and their resultant effect on grass carp stocking rate requirements (right)

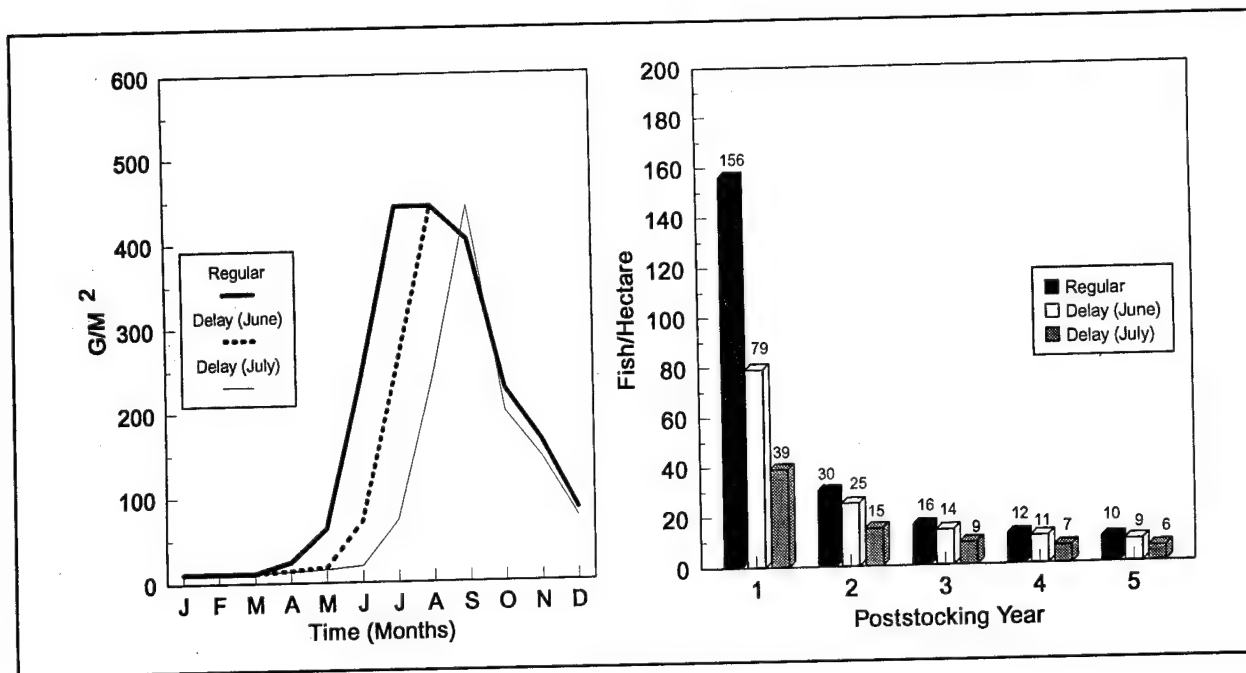


Figure 3. Seasonal plant biomass curves generated by the AMUR/STOCK model for differences in onset of plant regrowth (left) and their resultant effect on grass carp stocking rate requirements (right)

regrowth was delayed until June, peak biomass was attained during August. When regrowth did not begin until July, peak biomass levels did not occur until September.

Simulation outputs were generated to provide estimates of the stocking rates required to control plants for each growth curve described above. Stocking rate requirements were very sensitive to timing of regrowth onset and poststocking time. For each poststocking year, plants that initiate regrowth in April required significantly more fish for control. The difference was approximately fourfold in Year 1 and twofold in subsequent poststocking years. As shown in Figure 3, fish required for control of plants growing at the regular rate ranged from 156 fish/ha (Year 1) to 10 fish/ha (Year 5). For plants where growth was delayed until July, the stocking requirement ranged from 39 fish/ha (Year 1) to 6 fish/ha (Year 5).

Levels of Grass Carp Mortality

Differential mortality levels in stocked grass carp are a major source of variability in stocking effectiveness. Factors accounting for mortality levels include stocking size,

predator population levels, and commercial fishing pressure. To illustrate the impact of fish mortality on stocking rate requirements, simulations were generated for annual mortality levels of zero, 10, and 20 percent.

Simulation outputs of stocking rate requirements for these three grass carp mortality levels are shown in Figure 4. Outputs for "zero mortality" simulations indicate that stocking rate requirements will decrease annually over the first few years, then stabilize for the remainder of the 10-yr simulation period. Increases in grass carp mortality rates resulted in greater stocking rate requirements for each poststocking year. Peak effectiveness (i.e., minimum stocking requirements) for simulations which experienced grass carp mortality losses occurred in poststocking Years 5 to 6, after which stocking rate requirements gradually increased.

Summary of Results

A number of interacting factors account for variability associated with grass carp effectiveness following release. The effects of these factors therefore must be considered in order to determine proper stocking rates for

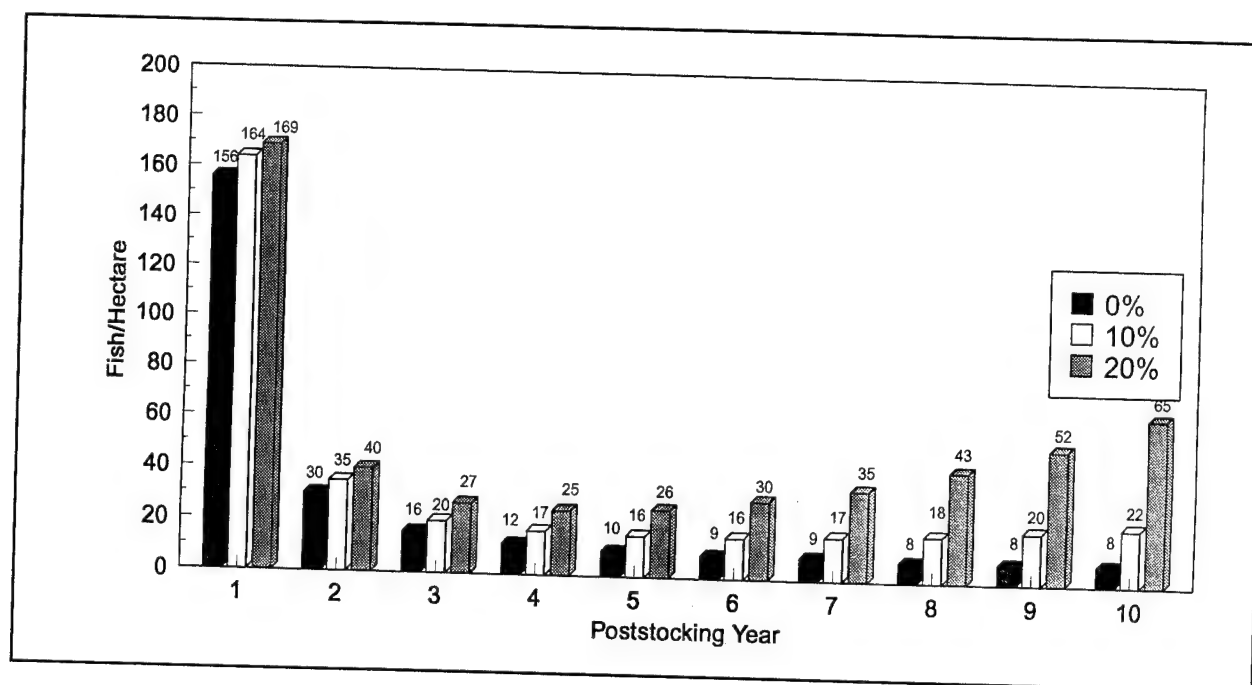


Figure 4. Effect of annual fish mortality on grass carp stocking rate requirements

the desired level of aquatic plant control. The WES Stocking Rate Model was designed to help aquatic plant managers consider the effects of some of these factors. Examples presented herein illustrate the significance of poststocking time coupled with three characteristics of plant growth (i.e. peak biomass level, overwintering biomass level, and timing of regrowth initiation) and with grass carp mortality rates.

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Conversion of HERBICIDE Version 1.0 for WINDOWS™ Applications

by

R. M. Stewart¹ and J. K. McAllister¹

HERBICIDE Overview

The **HERBICIDE Version 1.0** software package is a decision support tool designed to help aquatic plant managers select and implement effective herbicide application techniques for aquatic plant control operations. An overview of the model and supporting databases was presented by Stewart (1993), and examples of the model's intended use were provided in Stewart (1994).

Major assumptions incorporated in the design of **HERBICIDE** are the following. First, the model assumes that the level of mortality to a target plant infestation depends on the target plant species, the active ingredient of the herbicide, and the concentration and exposure time "dosage" resulting from the herbicide application.

Second, the model assumes that concentration/exposure time (CET) relationships developed for various combinations of aquatic plant species and herbicide active ingredients are accurate. With regard to postapplication aqueous concentrations of active ingredients, the model assumes that degradation and partitioning due to the processes listed in Table 1 will account for the majority of concentration reductions in the treatment site over time.

Further, the model assumes that the active ingredient will instantaneously achieve complete mixing within the treatment area water column following application, or following release from the formulation if a slow-release product. Partitioning within sediments and plant tissues is assumed to reach equilibrium at each time step and is based on partitioning coefficients and bioconcentration factors re-

Table 1
Major Fate Processes and User Inputs
to the HERBICIDE Model

Fate Process	Input Requirements
Transfer Processes	
Drift	Percent loss of active ingredient
Dilution	Application rate of formulation Percent active ingredient Release half-life of formulation Average depth of treated area Water exchange rate
Sorption	Herbicide sediment layer partition coefficient Total suspended solids Sedimentation rate Depth of active sediment layer Sediment water content, percent Sediment diffusion exchange rate
Volatilization	Volatilization half-life in water
Bioaccumulation	Bioaccumulation factor in plants
Transformation Processes	
Oxidation	Oxidation half-life in water Oxidation half-life in sediments
Hydrolysis	Hydrolysis half-life in water Hydrolysis half-life in sediments
Photolysis	Photolysis half-life in water Photolysis half-life in sediments
Biodegradation	Biodegradation half-life in water Biodegradation half-life in sediments

ported in the literature. Degradation processes are assumed to be accurately represented by half-life decay rates that are also available in the literature.

Finally, the model assumes that water exchange results in spatially uniform dilution of the herbicide active ingredient within the treatment area.

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

Conversion to WINDOWSTM

Consistent with the long-range goal of developing the APROPOS system described by Madsen (1995), simulation models developed for the APCRP have been converted to allow execution under the WINDOWS operating environment. In addition to providing accessibility through APROPOS, the WINDOWS versions offer several user-friendly enhancements. These enhancements for the **HERBICIDE** model are described below.

Main Menu

The Main Menu of the **HERBICIDE** model is illustrated in Figure 1. Through this menu, which appears upon execution of the model, the user is given access to the main toolbar, which provides icons for accessing the nine primary functions of the model.

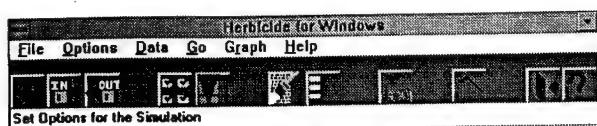


Figure 1. The Main Menu of the **HERBICIDE** model under the WINDOWS operating environment provides "point and click" control of model execution

Herbicide/plant combination icon

By double-clicking this icon, the user gains access to the menu illustrated in Figure 2. This menu allows the user to select from the avail-

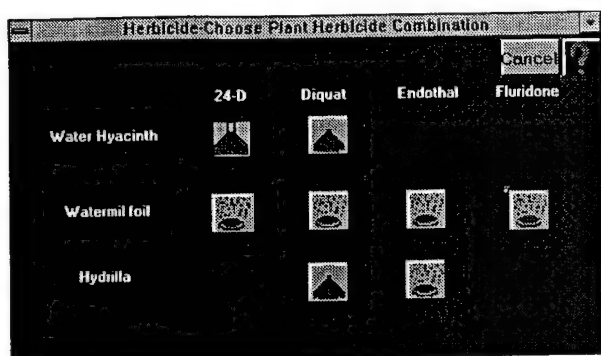


Figure 2. Menu from which the user selects the combination of herbicide formulation and plant species to consider in a simulation

able choices of herbicide formulation and plant species combinations for the simulation run. Selection of one of these choices allows the model to load default settings for the simulation run. These default settings are stored in a resident file specific for the selected herbicide and plant. After the default data file is uploaded, the user may modify any or all of the initialization settings by selecting the *Data Entry* icon discussed below.

Data Entry icon

Selecting this icon from the Main Menu activates the menu displayed in Figure 3. Through this active menu, the user is allowed to modify the contents of the initialization settings of the default data set selected from the menu illustrated in Figure 2. Data entry areas for each of the initialization parameters listed in Table 1 are accessible through this menu. For simulations in which the user chooses to modify default settings through this menu, the resulting "modified" data set can be stored for later retrieval and use (see icons *File Output* and *File Input* below).

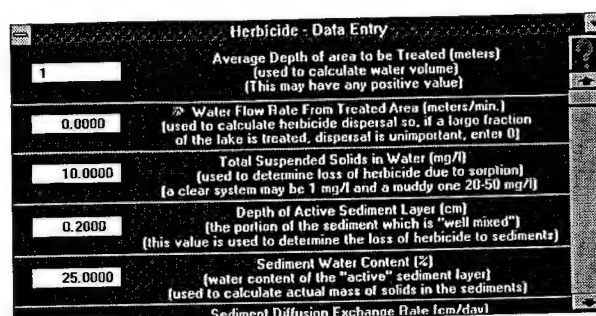


Figure 3. The Data Entry Menu allows the user to modify contents of the default initialization settings for the selected herbicide and plant

File Output icon

If the user modifies a default data file in any form, or entirely creates a model initialization data set to meet the needs of a simulation run, that data set can be stored for later retrieval and use by pressing the *File Output* icon. If this icon is selected, the model will prompt the user for a unique filename in which to store the contents of the data set.

File Input icon

Selecting this icon from the Main Menu toolbar allows the user to select and load a previously stored file containing a modified initialization data set.

Options icon

This icon provides access to the simulation run Options Menu illustrated in Figure 4. The user can select either metric or non-metric units of measure, as well as the type of output device. The user can also select whether to consider herbicide concentrations in the water column or in plant tissues. If water column concentrations are selected, and if a lower water concentration threshold is specified (see *Thresholds* icon), the user can specify how additional infusions will be added during the simulation to keep the water concentration above the threshold value. For simulations that consider slow-release herbicide formulations, the user is allowed to specify if the formulation release characteristic is exponential (that is, half-life release rate) or constant.

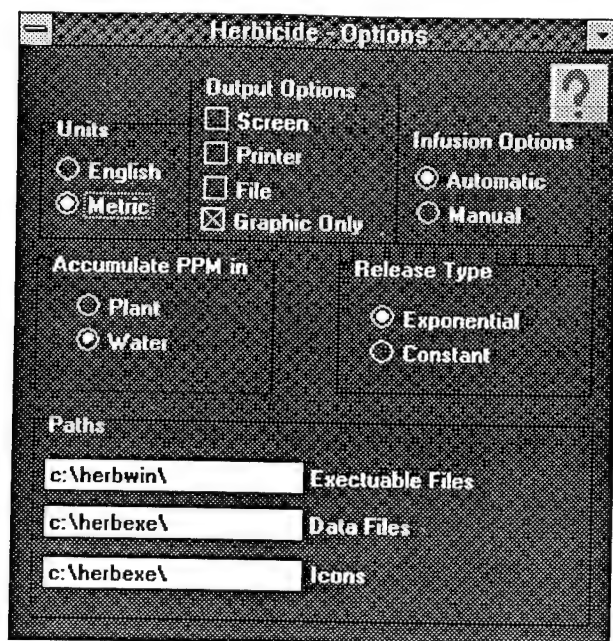


Figure 4. The HERBICIDE options Menu allows the user to define important "run-time" features of the simulation

Thresholds icon

The menu activated by this icon is illustrated in Figure 5. The user can enter the water threshold concentration of the herbicide active ingredient that elicits either an automatic infusion or a "user prompt" to accept a user-specified infusion. If the user does not change the default threshold setting of 0.0, then no additional infusions are considered in the simulation run. This menu also permits specifying the range (that is, maximum and minimum) of concentrations to be highlighted in the output graphs for the simulation. This highlighting helps the user visualize the length of time that the herbicide concentration remained within the specified target range. This feature helps the user compare model outputs with CET relationships developed for each combination of herbicide and target plant species.

Model Availability

The HERBICIDE model has been modified to operate under the WINDOWS operating environment and is currently available to users through the Aquatic Plant Control Bulletin Board System (APCBBS). The model will be distributed with a User's Manual at the end of the current fiscal year.

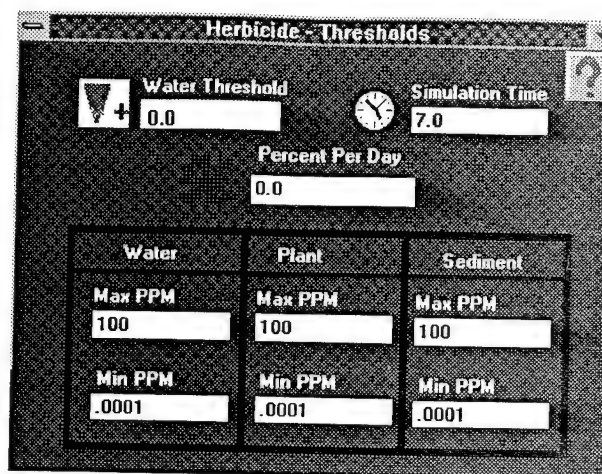


Figure 5. The Thresholds Menu allows the user to set upper and lower concentration thresholds that determine additional herbicide infusions

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- Madsen, J. D. (1995). See pp 283-285 in this proceedings.
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The Importance of Spatial Data in the AMUR Stocking Rate Model

by
E. M. Causey¹ and M. R. Kress¹

Introduction

The spatial distribution of aquatic plants in a water body is dependent upon key environmental conditions such as water depths and substrate conditions. It is important to understand and consider these spatial characteristics in numerical modeling and when developing control and management plans. This paper describes the use of spatial information for the WES-developed AMUR stocking rate model.

Several spatial elements are important to the Amur model calculations. These are:

- (1) Aquatic plants often occur in a complex of plant associations.
- (2) The plants occur across a range of environmental conditions especially variations in water depth.
- (3) Plants grow at different rates resulting in a wide range of plant biomass conditions.
- (4) Grass carp have different feeding preferences among plant species.
- (5) Plant competition allows new species to infest areas where existing species are controlled.

A method for considering each of these factors in AMUR calculations is presented below. AMUR fish requirement calculations for the 1989 Lake Guntersville conditions are compared with the actual 1990 fish releases in that water body.

Complex Plant Associations

The TVA delineated the 1989 aquatic plant distributions in Lake Guntersville from aerial photographs collected in the fall of that year. Data are presented here for only the area covered by TVA navigation charts 501, 502, and 503 (Figure 1). Figure 1 also shows the 1990 grass carp release sites and the number of fish released at each. The plant distribution data were digitized and input to a geographic information system (GIS) for geographic rectification and analysis.

Nine plant species were identified from the 1989 photography. These were mapped as single-species stands and in 32 different plant associations. Table 1 summarizes the aquatic plant population and shows the species and plant associations identified. The dominate species was *Myriophyllum spicatum* (MS). This species occurred alone and as a component of 22 types of mixed stands. Figure 2 illustrates the plant distribution patterns in Lake Guntersville.

To efficiently use this plant distribution information during numerical modeling, the area of individual species was determined. The total area of species mapped in single-species stands was determined. To estimate the area represented by each species in a complex plant association, weighting factors were assigned. In this example only the first two species in the association were considered. The first species was assigned a weighting

¹ U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

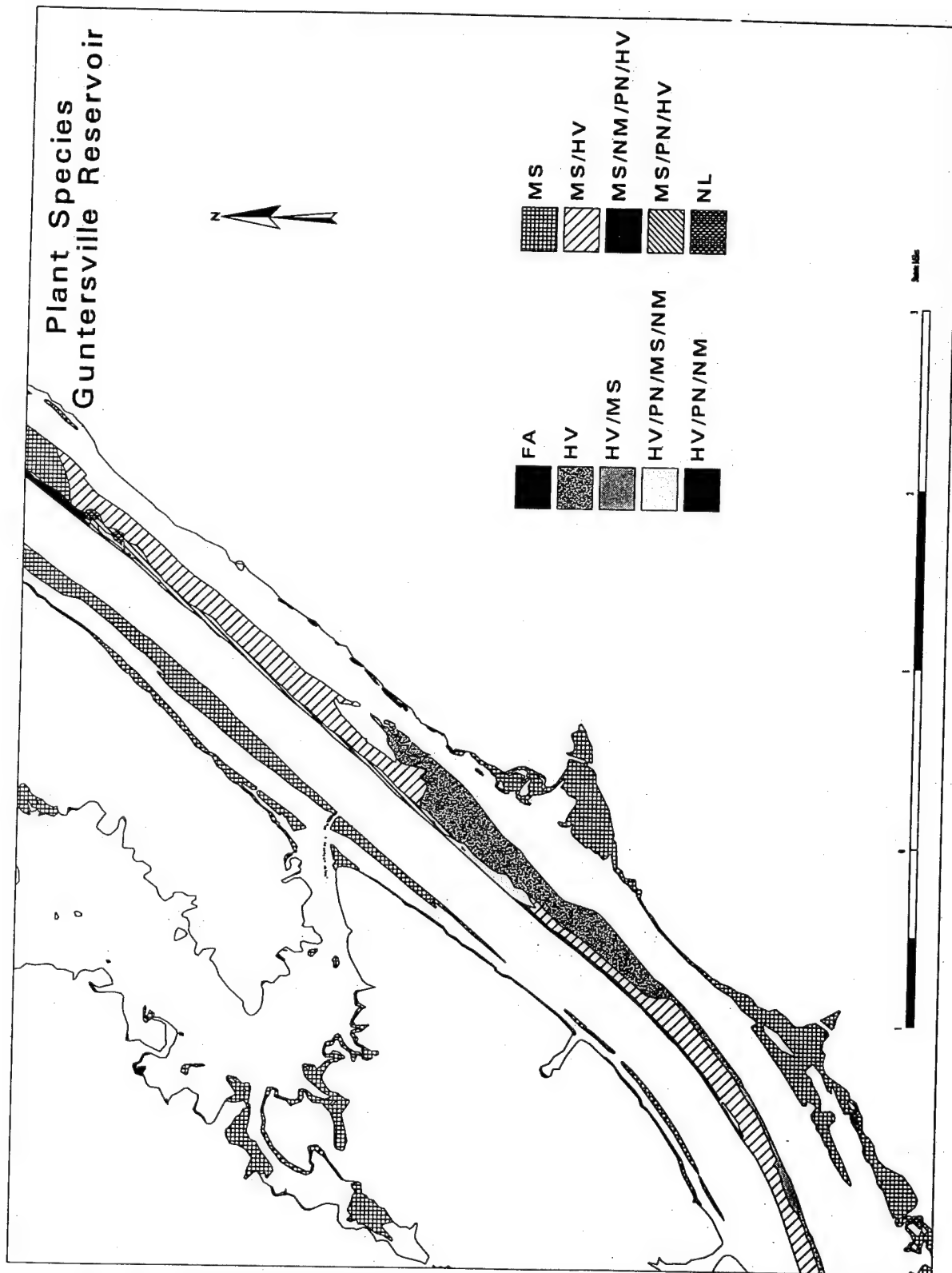


Figure 2. An illustration of the 1989 plant distribution for an area of Lake Guntersville

Table 1
Aquatic Plant Combinations Delineated
from 1989 Aerial Photography of
Guntersville Lake

Single Species Stands

Cabomba caroliniana (CC)
 Ceratophyllum demersum (CD)
 Chara sp. (CH)
 Elodea canadensis (EC)
 filamentous algae (FA)
 Hydrilla verticillata (HV)
 Myriophyllum spicatum (MS)
 Nelumbo lutea (NL)
 Najas minor (NM)

Mixed Species Stands

CD/MS
 CD/MS/NM
 CD/NG/NM/MS
 CD/NM
 CD/NM/MS

 CH/FA
 CH/MS
 CH/MS/HV
 CH/NM
 CH/NM/NG

 FA/MS

 HV/MS
 HV/PN/MS/NM
 HV/PN/NM

 MS/CD
 MS/CD/NG/PN
 MS/CD/NM
 MS/CH
 MS/FA
 MS/HV
 MS/HV/CD
 MS/NM
 MS/NM/PN
 MS/NM/PN/HV
 MS/PN
 MS/PN/HV
 MS/PN/NM

 NM/CD
 NM/CH
 NM/NG
 NM/PN
 PN/NM

factor of 0.65 and the second species a weighting factor of 0.35. For example, a 100-ha stand identified as HV/MS was estimated to contain 65 ha of HV and 35 ha of MS.

Water Depth Variations

In the AMUR model plant growth rates and predicted plant biomass are a function of water depth. Water depth information in Lake Guntersville was included in the GIS database and water depth was mapped in 1.5-m increments based on the bathymetric data shown on the TVA navigation charts. Figure 3 shows a portion of the water depth data.

An analysis of water depth and plant types was conducted using the GIS. The species weighting factors discussed above were applied during this analysis. The area of plants by species and water depth are listed in Table 2. Plants other than HV and MS were combined and referred to as annuals.

Table 2
Area of Aquatic Plants by Water Depth,
Lake Guntersville (Nav Charts 501-503,
1989)

Water Depth, m	Area, ha			
	Hydrilla	Annuals	Milfoil	Total
0-1.5	49	161	1,376	1,586
>1.5-3.0	205	47	1,377	1,629
>3.0-4.5	233	6	410	649
Total	487	214	3,163	3,864

Variable Plant Growth Rates

The AMUR stocking rate model provides estimates for the number of fish per hectare of infestation that must be stocked in a given month to control a specified type and density of aquatic plants during the first year. Table 3 contains the AMUR model estimates for the nine plant type-water depth categories. The predicted August plant biomass values in

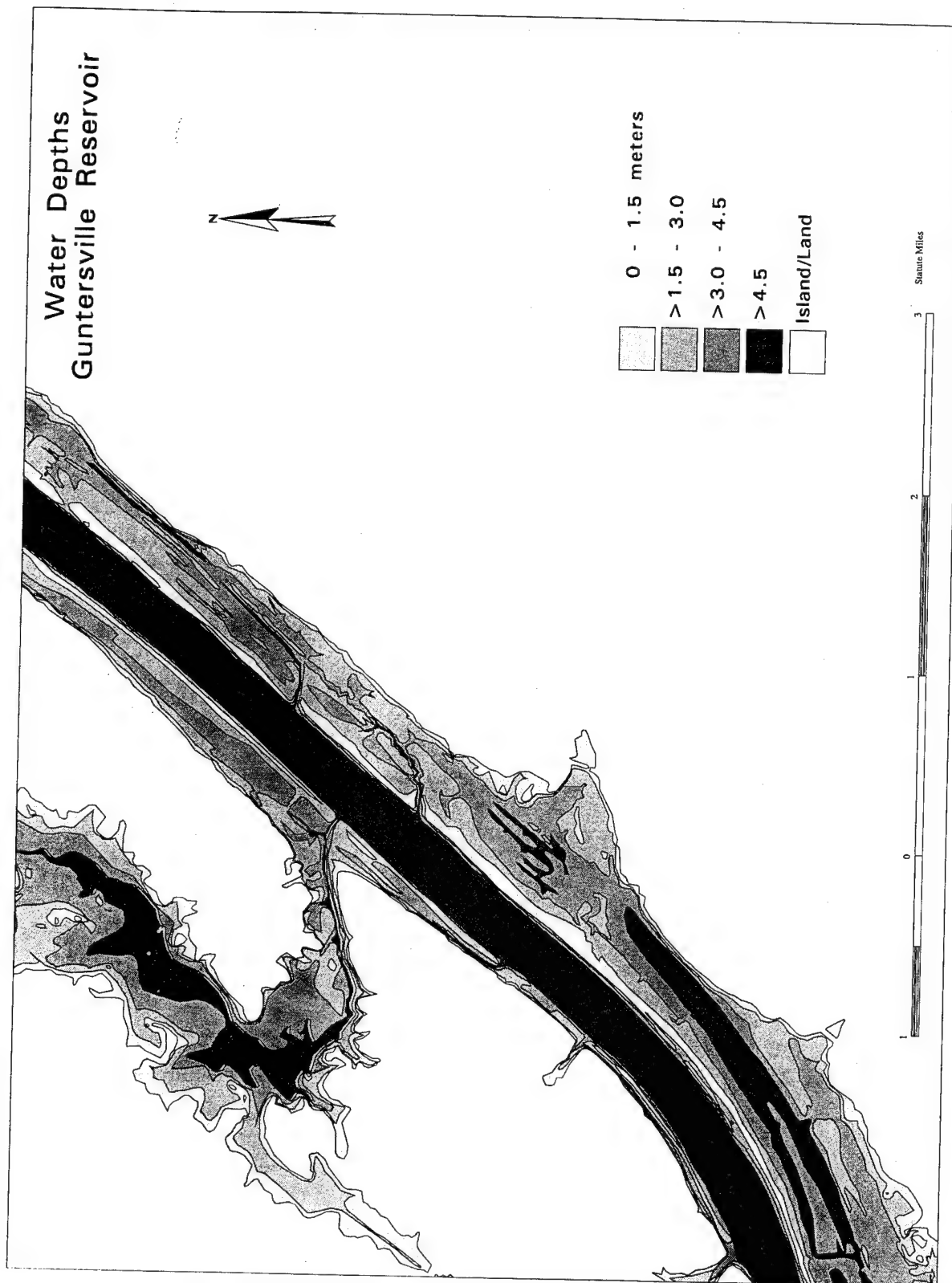


Figure 3. Lake Guntersville water depths for same area as plant distribution shown in Figure 2

Table 3
Predicted August Plant Biomass (kg/m²)
and Fish Required for Control by Year
One (fish/hectare)

Water Depth, m	Biomass/Fish		
	Hydrilla	Annals	Milfoil
0-1.5	4.51/222	1.25/59	1.61/164
>1.5-3.0	4.51/167	1.25/24	1.61/74
>3.0-4.5	2.25/71	1.26/24	1.16/27

Table 3 are those estimated by AMUR for Lake Guntersville environmental conditions assuming no grass carp had been stocked. The fish/hectare values are the number of fish estimated that would be required to control the plants at the specified depth in the first year.

The difference in the number of fish needed for similar biomass values in deeper

water reflects a slower plant growth rate. Monthly biomass (kg/m², fresh weight) estimated by AMUR for three plant types and three water depths in Lake Guntersville are listed in Table 4. Uncontrolled HV growing in 2.0 m of water could eventually reach the same biomass as that in 1.0 m of water (Table 4). However, the rate of growth in deeper water is slower until July. This reduced growth rate in the first half of the year allows fewer carp to affect control in those areas.

Total Fish Stocking Requirements

The number of fish required to control the 1989 aquatic plant population in Lake Guntersville, as estimated by AMUR, are listed in Table 5. Fish required for control in each plant - water depth combination were

Table 4
Monthly Biomass (kg/m² Fresh Weight) Estimated by AMUR for Three Plant Types
and Three Water Depths

Month	Hydrilla			Annals			Milfoil		
	Water Depth, m			Water Depth, m			Water Depth, m		
	<= 1.5	>1.5-3.0	>3.0-4.5	<= 1.5	>1.5-3.0	>3.0-4.5	<= 1.5	>1.5-3.0	>3.0-4.5
Jan	0.45	0.11	0.11	0.05	0.05	0.05	0.08	0.08	0.08
Feb	0.45	0.11	0.11	0.05	0.05	0.05	0.08	0.08	0.08
Mar	0.45	0.11	0.11	0.05	0.05	0.05	0.08	0.08	0.08
Apr	0.90	0.24	0.13	0.10	0.06	0.06	0.16	0.09	0.09
May	2.39	0.65	0.14	0.27	0.07	0.07	0.43	0.10	0.09
Jun	4.51	2.47	0.55	1.05	0.26	0.26	1.61	0.39	0.10
Jul	4.51	4.51	1.96	1.25	0.92	0.92	1.61	1.40	0.35
Aug	4.51	4.51	2.25	1.25	1.26	1.26	1.61	1.61	1.16
Sep	4.11	4.10	2.05	1.14	1.15	1.15	1.47	1.47	1.61
Oct	2.31	2.30	1.15	0.64	0.64	0.64	0.82	0.82	0.73
Nov	1.68	1.68	0.84	0.47	0.47	0.47	0.60	0.60	0.53
Dec	0.86	0.86	0.43	0.24	0.24	0.24	0.31	0.31	0.27

Table 5
Area of Plants by Water Depth and Fish Required to Control in Year One (Nav Charts 501-503, 1989)

Depth, m	Hydrilla, ha	Fish	Annuals, ha	Fish	Milfoil, ha	Fish	Total, ha	Total, fish
0-1.5	49	10,878	161	9,499	1,376	225,664	1,586	246,041
>1.5-3.0	205	34,235	47	1,128	1,377	101,898	1,629	137,261
>3.0-4.5	233	16,543	6	144	410	11,070	649	27,757
Total	487	61,656	214	10,771	3,163	338,632	3,864	411,059

determined as the plant area (hectare) times the respective fish/hectare value calculated by AMUR (from Table 3).

A total of 100,000 diploid grass carp were released by TVA into Lake Guntersville between 26 April and 31 July 1990. The releases were widely distributed throughout the lake as shown in Table 6 and in Figure 1.

Table 6
Fish Released (26 April - 31 July 1990)

Nav Chart	Fish
501	29,000
502	33,050
503	37,950
Total	100,000

Plant Feeding Preferences

To estimate the level of control possible by 100,000 fish, their distribution pattern within the plant-infested areas must be specified. For example, if the 100,000 fish are evenly distributed throughout the plant-infested areas and do not exhibit a preferred feeding preference, the resulting fish density would be approximately 26 fish/hectare. Table 3 shows that this number of fish the first year is only sufficient to control annuals in water greater than 1.5 m. Under an even distribution, no preference assumption, the 100,000 fish would not provide control for any of the HV- or MS-infested areas.

Evidence (Leslie et al. 1987) suggests that grass carp exhibit distinct feeding preferences. The order of preference for the plants in Lake Guntersville is first HV, then annuals, and finally MS. In Table 7, the feeding preference is applied in estimating the level of control provided by 100,000 fish. According to the feeding preference, the number of fish feeding on hydrilla are estimated to be 61,656 fish (see Table 5). The next preference by the fish are the annuals and thirdly the preference is milfoil (MS). When feeding preferences are included, the 100,000 stocked fish are estimated to be sufficient to control all HV, annuals, MS in 3.0-m water and 223 ha of MS in 1.5-3.0 m.

Table 7
Predicted Control Year One (Nav Chart 501-503, 1990)

100,000	Fish Stocked	Feeding Preference
-61,656	Hydrilla control	High
-10,771	Annuals control	
27,573	Available to feed on milfoil	Low
-11,070	MS (>3.0-4.5 m) control	
16,503	Available to feed on milfoil (74 f/ha)	
-16,503	233 ha MS (1.5-3.0 m) control	
0		

Conclusion

Many of the environmental and biologic factors influencing the success of aquatic plant control programs have spatial components which can be quantified and used with the aquatic plant control numerical models. Spatial data and analysis tools are being used in combination with AMUR and other simulation models to assist in evaluating different management alternatives.

Reference

Leslie, A. J., Jr., Van Dyke, J. M., Hestand, R. S., and Thompson, B. Z. (1987).

"Management of aquatic plants in multi-use lakes with grass carp (*Ctenoparyngodon idella*).” Lake and reservoir management: Vol II. *Proceedings, 5th Annual Conference and International Symposium*. G. Redfield, J. F. Taggart, and L. M. Moore (eds.), North American Lake Management Society, Washington, DC, 266-76.

Aquatic Plant Management Strategy Planner

by

John D. Madsen¹ and Joseph K. McAllister, Jr.²

Almost 30 years of research and development within the Corps, combined with active research and development by other Federal and state agencies, universities, and commercial enterprises, have produced a vast body of largely unintegrated aquatic plant management information. An integrated systematic method of retrieving this information is needed. Recent developments in personal computer software and hardware have made it feasible to develop a systematic method of information access and retrieval that the majority of Corps plant managers can utilize. An aquatic plant management strategy planning tool built with this expanding technology is needed for the Corps of Engineers.

The development of the APROPOS System (Aquatic Plant Resource Operations & Planning Online Support) addresses this need. APROPOS is a computer-assisted tool that will facilitate consideration of available information for the development of management plans. APROPOS will have a GUI (graphics user interface) shell which accesses modules to develop systematic aquatic plant management plans, retrieve necessary information, operate computer simulations, utilize databases, and interface with geographic information systems (GIS). During its development, the APROPOS system will also illuminate further areas for research.

The main menu of the APROPOS will allow access to the planner, literature databases, simulation tools, field technique toolbox, control technique toolbox, database menu, and help menu, as is typically found in Windows® (all registered tradenames are the property of their respective owners) programs (Figure 1; Table 1). The APROPOS system will be mod-

ular to allow upgrading information and tools, allow components to be used as stand-alone packages as desired, and ensure that components can be incorporated into the system as they become operational.

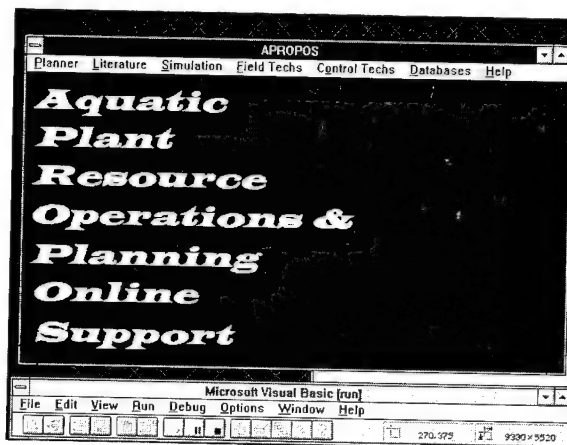


Figure 1. Main menu for APROPOS

The planner section contains the tools necessary to create an operational, site-specific management plan (Figure 2). The components require that this process initiate with identifying the problems of a given site, the interest groups which may be involved, the management goals, and potential solutions to achieve the management goals. Since planning is spatially oriented and site-specific, a sketchpad for non-GIS users is included in this section. The sketchpad can create rough diagrams of the management site that contain features relevant to management planning. Additional tools will assist in developing a planning and operational timeline, maintaining a record of contacts with interest groups, identifying financial and human resources available to the operational plan, and verifying the efficacy of the plan.

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² Computer Sciences Corporation, U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.

Table 1
Main Menu Components of the APROPOS System

Main Menu	Components	Purpose
Planner	<ul style="list-style-type: none"> • Situation • Report • Sketchpad 	<ul style="list-style-type: none"> • Builds, modifies, prints, and evaluates plans
Literature	<ul style="list-style-type: none"> • Bibliographic database • Modem access to other sources • Information on supplementary databases • Directory of personnel 	<ul style="list-style-type: none"> • Searches for key information sources, direction to additional help, access to some reports and abstracts
Simulation Toolbox	<ul style="list-style-type: none"> • Plant growth • Control techniques • Environmental effects • Economic 	<ul style="list-style-type: none"> • Allows examination of potential impacts of selected actions.
Field Techniques Toolbox	<ul style="list-style-type: none"> • Hypertext manuals • Expert systems • Plant identification guide 	<ul style="list-style-type: none"> • Presents guides on developing sampling plans and carrying out data collection
Control Techniques Toolbox	<ul style="list-style-type: none"> • Biological control • Chemical control • Mechanical control • Physical control • Alternative controls 	<ul style="list-style-type: none"> • Accesses critical information on each control technique and its alternatives
Database Toolbox	<ul style="list-style-type: none"> • Case studies • GIS access (ArcView) • Other databases, database management tools • Straightforward statistical analysis 	<ul style="list-style-type: none"> • Reviews how other managers have approached similar situations • Accesses database
Help Function	<ul style="list-style-type: none"> • Context-sensitive hypertext tutorial 	<ul style="list-style-type: none"> • Assists user in operation of program

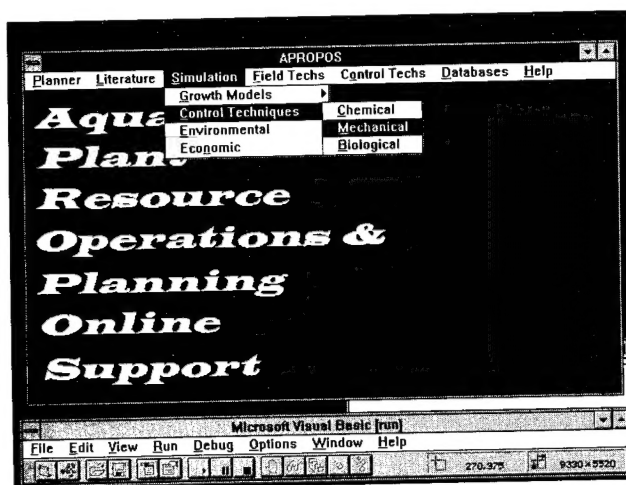


Figure 2. Planner submenu for APROPOS

The literature database will include key reference materials, including those used in the development of other toolboxes. Some citations

will include abstracts or entire sections of text for further study. The sources for these references will include scientific and technical reports and publications, operational information, regulatory guides and statutes, herbicide labels, and environmental information.

The simulation toolbox will provide access to simple models to project the potential results of different management alternatives. The submenus include plant growth models, control technique simulations, environmental simulations, and economic simulations. Models will include those produced by the Aquatic Plant Simulation Team, other teams within WES, researchers outside of WES, and potential future collaborators.

The field techniques toolbox will assist in developing a sampling program best suited to evaluate the objectives of the management

plan, with a minimal amount of cost. The field techniques toolbox will give the manager a working knowledge of various monitoring techniques so that they can collect the data themselves or oversee the efforts of others. Fundamental to this effort will be identification of both target and native plant species through a plant identification "expert system" included in the field techniques toolbox. Quantitative methods for discrete population sampling will include cover estimation, transect, and biomass techniques. For larger scale evaluations, GPS, bathymetric, and remote sensing techniques will be presented. Other sampling techniques, such as evaluating water movement and herbicide dissipation, will also be explained. Data entry, analysis, and evaluation can be performed within the database menu. Since identical database structures will be used, data can be shared between different users.

The control techniques toolbox will access information needed for selecting between different management techniques, based on economic, environmental, and efficacy concerns. The control toolbox will access information on biological, chemical, mechanical, and physical control techniques, as well as less traditional control techniques.

Four independent submenus are found under the database heading: case studies, GIS, other databases, and data analysis (Figure 3). These four semi-independent components interact to form a powerful tool to view past, present, and future data.

Case studies will report both successes and failures in aquatic plant management and provide insights into the causes of these outcomes. Ideally, the manager will be able to see how others approached a similar situation and their results. As more individuals use the APROPOS report functions, an expanding database will allow managers to get an overview of how aquatic plants are being managed nationwide.

The GIS interface will initially allow those with ArcView[®] and an existing GIS to interactively explore their data while within APROPOS, and transfer data from the GIS to the

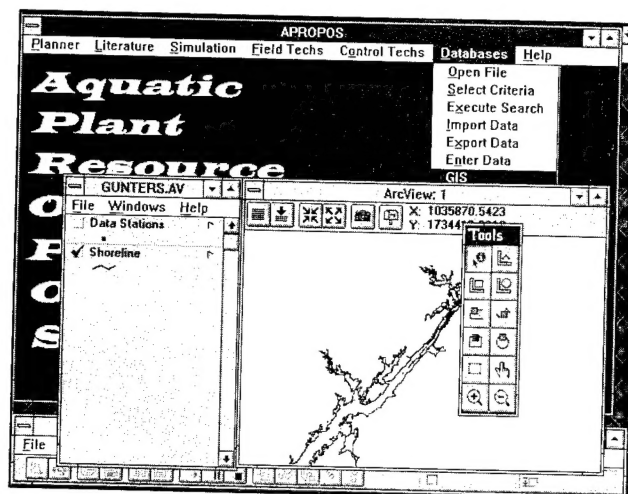


Figure 3. Database submenu for APROPOS, with window to ArcView[®]

planning modules (Figure 3). Eventually a more direct link to ArcInfo[®] will be made, which will allow two-directional flow of data and viewing of simulation results.

Other databases will also be accessible through the database menu, the most important of which will be the site-specific information collected and entered by the manager. Since the format will be set and standardized, these data can be easily exchanged and utilized for case studies by other APROPOS users. Databases generated for simulation model initializations will also be available from the Aquatic Plant Bulletin Board Service (APBBS).

A simplified data analysis program will allow statistical analysis and graphical presentation of data from different sources such as data collected by the manager, case studies, and simulation output visualization. The graphical presentation tool is already under development within the Simulation Team. Software routines to link APROPOS to popular software programs are also under consideration.

A context-sensitive help function will assist the user in navigating through the APROPOS functions. For more information on related topics, the user will click on highlighted text ("hypertext") to access additional information. These functions will allow users to utilize the program without reading a manual or knowing a given command. Hypertext manuals will

also be used throughout the various toolboxes and menus to allow the user to rapidly access and cross-reference needed information.

APROPOS will be readily upgradable because of its modular design. Many components will be usable as stand-alone features even before the entire system is completed. Its flexibility, expandability, and growth potential are almost limitless. In addition, the user-friendly interface ("Windows") will make it an approachable technology transfer tool.

To be a success, APROPOS will need input from WES researchers, field personnel, state cooperators, and other interested individuals. During tool development, the assistance of WES technology teams will be sought for development, review, and updates on key

components. Feedback from the Field Review Group and potential users will also be solicited through workshops and beta/alpha evaluation releases of the system. In addition, anyone interested in commenting on the APROPOS system may do so through questionnaires on the APBBS, or they may send general comments to the authors using the APBBS message service. The first questionnaire for APROPOS feedback is online now.

The APROPOS system will allow the manager to easily access information necessary to make a comprehensive aquatic plant management plan. The system will also provide a systematic planning framework and develop a consistent reporting format for interchangeable databases for aquatic plant management projects. The goal is not only to make aquatic plant management better, but easier.

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